

nature

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The cost of Swiss francs

It is fashionable to see the late 1960s as a period when science on a worldwide basis went through a major transition, emerging with a much-changed relationship with society in general. That transition, even if major, was at least fairly gradual and allowed for steady adjustment in values amongst scientists. In later years we may well look at 1976 and decide that here was a second equally drastic transition, but one which occurred in the course of weeks rather than years and which affected only Britain. The continuing weakness of the pound—a result not of irresponsible financial journalism, nor even of sterling's role as a reserve currency, but of long-term industrial inadequacy to compete successfully in overseas or even domestic markets—is precipitating a serious crisis of confidence among many scientists about the will, let alone the ability, of Britain to continue as a pre-eminent scientific nation. Much of this alarm is exaggerated, there is no doubt, and there is a danger that we all think ourselves into a worse crisis than really exists.

The details barely need repeating now. Every cent that the pound drops on the foreign exchange markets adds nearly £200,000 to the money that has to be found by the Science Research Council (SRC) to pay international subscriptions, notably to CERN, the European Space Agency, and the Institut von Laue-Langevin. The CERN subscription, at over £20 million, is by far the greatest. The recent downward trend of the pound has thus meant severe economies for domestic programmes in no way related to high-energy physics, even those of other research councils. It has also meant that Britain has had to put out some very careful feelers to CERN and its member states for sympathetic understanding.

What are the options at Geneva? It would conceivably be possible to pull out of specific projects there by way of economy. There has been recent speculation about dropping out of the Intersecting Storage Rings programme, which provides collisions at the highest energy in the world. But there seems no evidence at all that Britain has either taken or intends to take any such step, which would be the most ham-fisted way of disrupting CERN's operations. The danger is that such speculation,

especially in an international framework, can easily turn into a self-fulfilling prophecy. It is much more likely that SRC, if it fails to convince the Treasury that support for an international treaty should be detached from a national policy on cash limits, will go to CERN to see if there is any way that the UK can reduce its overall contribution while maintaining its membership of all programmes. It goes without saying that to have to do this would hardly enhance our reputation as a partner in European ventures.

It is difficult as yet to predict how our partners in CERN would react to such an approach. No other country seems to be experiencing similar problems; Italy might seem the most vulnerable but pays directly from its Foreign Ministry, which has been in the habit of paying without question. One suspects, however, that there will be genuine support, provided that this is not to be an annual affair. The only difficulty is that the Germans, who might logically be bearers of a greater burden, already contribute 25% of CERN's budget, which is the limit that any country may carry. Thus any load shed by Britain could possibly prove an embarrassment for a smaller country to pick up.

But where in all this is the turning point for British science? Will this not seem a relatively minor ripple in hindsight? There are grounds for thinking otherwise, that the affair could polarise the scientific community to its great detriment. There has, of course, for some time been a substantial "anti-big science" feeling only just below the surface in many sectors of the community; the events of the past few weeks with their resultant stringencies across the board can hardly have helped dissipate this feeling. On the other hand the high-energy physicists now make no secret of dissatisfaction with the way SRC plans to shift emphasis in its domestic funding. In particular many are expressing a new-found concern at the council's growing emphasis on engineering research.

Whatever the merits of either of these points of view, they are symptomatic of dangerous splits and uncreative tensions which could very easily harm Britain's research potential and lead her down a depressing road to second-rate status. □



The loneliness of the original investigator

Ernest Borek of the University of Colorado Medical Center comments on the fate of original research under present funding arrangements

THE curse on science is that its masters demand results they can readily comprehend. Kings and princes once gave what we would call today research grants to their alchemists, expecting gold for their largesse. The alchemist Hennig Brandt serendipitously discovered that if he performed some hocus-pocus on dried urine, he obtained a magic stuff. It glowed in the dark! His patron, a German prince, was not impressed, so Brandt's grants continued to be miserly.

Of course, Brandt discovered phosphorous, and our knowledge of the universal need for this element in the form of phosphates by plants and the consequent incorporation of it into fertilizers has improved the yield of crops, producing wealth over the scores of years, which in value probably exceeded all the gold of the world.

Our situation today is not much different. We are the remittance men of Presidents and Royal Commissions. Science is in the political arena; we are at the mercy of the ringmasters. Charles G. Wilson, a Secretary of Defense whose department controlled enormous funds for research, pithily (if ungrammatically) aphorised the opinion of many upper level administrators on research: "Basic research is when you don't know what you're doing".

Five years ago the President of the United States decided, it being a pre-election year, to sign the "Conquest of Cancer Act of 1971", and did so with a flourish. He invited a horde of television men, some legislators, and a sprinkling of scientists to witness the great moment. I am ashamed to admit that curiosity impelled me to accept the invitation. In those pre-Watergate days, I was almost touched when the president told us about his dear aunt who died of cancer. In less than a year he impounded a hundred million dollars from cancer research provided by the bill he signed with so much emotion.

The British scientist is also not free from the paralysing shackles of authority. A few months ago, a young Englishman spent a year in my laboratory learning the skills to study the modifications of tRNA. He went home filled with plans and enthusiasm. He actually made an observation which could have been developed into something very interesting. But soon I received a sad letter from him. The

director of his research institute ordered him to drop studying modification of tRNA because the molecular biologist on some high MRC committee put a very low priority on such research. Chalones were in vogue.

Now this is hearsay and may be just sour grapes, but I could count on the fingers of one hand the British colleagues contributing visibly to this field. For the benefit of those who are not molecular biologists, I should point out that the primary structure of tRNA is made by one enzyme, but more than fifty modifications of it are achieved by perhaps 150 different enzymes. We know the functions of some of these modifications, but for most of them, the gold is not yet visible. tRNA is the most versatile of the biomacromolecules. In addition to its pivotal participation in protein synthesis, it can regulate transcription of DNA, translation of messenger RNA and can even control enzyme activity. What new roles will be revealed by patient probings cannot be predicted.

The American scientist who strives for originality is bedevilled at two levels of the governance of science. The General Accounting Office (GAO), which is the administration's watchdog over the budget, has recently urged Congress to require annual reviews of all research grants awarded by the National Institutes of Health (NIH). One can just see the staff at GAO with their tidy accountant's minds fretting over the possibility that some scientist may goof off for three years. After all, the corporations from where they came give quarterly reports of their earnings to the stockholders.

Unfortunately, at NIH the administrators urge the appointment of young investigators to peer review groups. James Watson is the most vocal champion of youth on advisory bodies because, he says, creativity is exclusively in the provenance of youth. This may be true in mathematics, but has no factual basis in the biological sciences. A young scientist becomes visible in biological research at an early age not by original major contributions but by frenetic publishing in a field opened by the originality of others. Polish a little and publish a lot, that is the leitmotif of their research careers. "Peers" like that cannot possibly appreciate a new idea or a new approach. At the end of a two- or a three-year grant period,

they measure accomplishment by counting pages rather than by the increment in our knowledge. The grant application of one of our most distinguished cell biologists went unfunded recently; the reason, he was told, was that his proposed experiments were too bold.

The members of advisory panels at the National Science Foundation (NSF) tend to be more mature and accomplished. But Congress insists that grants be awarded for only two years. How can one pose a challenging question and dare explore it when within a year it is time to write the grant renewal? At a recent conference on nucleic acid modification I noted that pre-eminence in the technology of tRNA modification has passed from America to Germany and Japan, and the reason must be that no one in his right mind would dare tackle such a problem on a two-year NSF grant (*Nature*, September 9, 1976).

Some 25 years ago I became engrossed in Lwoff's discovery of lyso-genic induction. In four years, we filled notebooks with observations I did not think were worth publishing. Had I depended on our current system of research support, I would have been washed out. Yet it is a matter of record that from those four years of observation came the discovery of "relaxed control" of RNA synthesis, indirect induction of latent virus, post-synthetic modification of nucleic acids, and the first qualitative difference in a biochemical component of a tumor versus a normal cell.

To beat the system some investigators resort to artful dodging. You describe in your grant application your second level ideas, or experiments you have already done and thus gain time and resources to bootleg explorations of your pet ideas. Five years ago I foolishly described, among other things, a programme of study in tissue slices or *in vitro* of the functional effects of hormone-induced modifications of tRNA. The critique relayed to me was that I had no experience in that field and the funding was proportionately reduced. Needless to say, I went on to do what I wanted (see *J. Biol. Chem.*, **248**, 762; 1973, and *Nature*, **262**, 62; 1976). During these bootlegging operations, we observed serendipitously a novel attribute of the carcinogen ethionine (see *Nature*, **259**, 588; 1976).

Many years ago while an undergraduate, I supported myself and my family as a waiter in a restaurant where one of my duties was selling bootleg whiskey. Now I bootleg my research. *Plus ça change* . . .

Whither US science now?

Colin Norman surveys the post-election scene from Washington

WHEN it became clear shortly before dawn last Wednesday that Jimmy Carter had the electoral votes needed to put him into the White House, many Americans switched off their television sets and went to bed knowing who their next President would be, but not what he will do. Substantive policy matters were almost submerged in a flood of trivia during the long, tedious election campaign as the candidates' many rhetorical blunders, racist remarks of Cabinet members and non-political matters such as abortion claimed the spotlight. Although the campaign lasted many months and cost millions of dollars, it seems to have left most voters puzzled about where the candidates stand on the major issues.

Nevertheless, it is evident that Carter has offered the American people a very different political philosophy from the one which prevailed during the Nixon and Ford Administrations. As the Carter Administration takes shape over the next few months, it is likely to chart new courses in areas such as health policy, taxation, nuclear power, environmental protection and government reorganisation.

Little part for science

Needless to say, scientific issues played little part in the election campaign. Science policy is scarcely a partisan matter, and it hasn't figured prominently in election rhetoric since the Kennedy campaign, which took place in the post-Sputnik panic of the late 1950s. But when Carter takes office next January, he will be confronted with a number of important decisions which will have direct impact on science and technology.

First, however, it should be noted that Carter will enter the White House with at least lukewarm support from the scientific community. Though few scientific groups campaigned actively in support of Carter—in previous elections, groups of scientists campaigned for Kennedy, Johnson and McGovern—opinion polls taken among American academics found that Carter was favoured by a margin of two to one. A poll taken for the *Chronicle of Higher Education*, for example, found that 54% of the scientists surveyed favoured Carter, compared with 22% for Ford.

Carter's first chance to strengthen (or, indeed, to disrupt) his links with the scientific community will probably be in his appointment of people to fill

top science posts in his new Administration. Nearly 3,000 government posts are filled by Presidential appointment, and thus a change of President is usually accompanied by a wholesale change of top-level bureaucrats.

It is worth noting that Richard Nixon amply demonstrated the potential for souring relations with the scientific community in such matters. Early in his Presidency, Nixon vetoed the selection of Franklin Long as Director of the National Science Foundation because of Long's opposition to the anti-ballistic missile programme. Though the veto was made before Long's selection was announced, word filtered out and a shrill outcry was raised against political screening for that normally apolitical post. And Nixon incurred the wrath of many scientists four years later when he sacked the head of the National Institutes of Health, allegedly for disagreeing with White House policy on biomedical research.

Thus, Carter will probably tread carefully. His chief selection will be somebody to fill the post of science adviser and director of the newly-created White House Office of Science and Technology Policy (OSTP). Since OSTP was established by legislation, Carter is bound to retain it—indeed, he has already said that his science adviser will "provide a permanent and high-level relationship between the White House and the scientific community". The present incumbent, Guyford Stever, said when he was appointed last August that he would not stay long after the election, no matter who won. A widely-tipped candidate for the post is Lewis Branscomb, former director of the National Bureau of Standards and now head of research at IBM. Branscomb coordinated a science policy task force which advised Carter during the campaign.

The other chief science posts that Carter must fill are Director of the National Science Foundation and Director of the National Cancer Institute. The NSF post was vacated by Stever when he joined the White House staff and the NCI job was vacated in September by Frank Rauscher, who took a more lucrative job at the American Cancer Society. Carter is also expected to replace Russell Train as head of the Environmental Protection Agency. Though Train is generally considered to have

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Ford: departure leaves vacancies

done a good job, the post is highly political and Carter made considerable mileage from attacking the Ford Administration's environmental record. On the other hand, Carter is expected to retain Donald Fredrickson as director of the National Institutes of Health. A skilled administrator, Fredrickson has earned a solid reputation and Carter is unlikely to risk provoking an outcry by replacing him for political reasons.

Carter will also inherit a top-level committee, chaired by Simon Ramo of TRW Inc, which was established by Congress to conduct a two-year investigation of federal science policy. The committee is scheduled to make its report and recommendations late in 1978, at which time Carter will have the option of retaining it as a senior White House advisory committee, or scrapping it.

Key policy decisions

As for the Carter Administration's likely policies for research and development, little is known with certainty, but many key policy decisions will have to be made in the

first few months after President Carter takes office. The following are some of the principal issues which Carter must decide.

● **Energy policy.** One area in which Carter has frequently attacked the Ford Administration's record is energy policy in general, and nuclear policy in particular. He has offered some specific plans for reorganising the federal energy bureaucracy, and for curbing nuclear proliferation.

Throughout the campaign, Carter made much of the fact that he was trained as a nuclear engineer, and therefore he claims to understand the problems associated with nuclear power. His attitude toward nuclear energy is decidedly cool, promising to minimise the United States' dependence on that source of power, but specifically stating that he does not support a nuclear moratorium. In particular, he has said that he will channel more energy research and development funds into alternative sources such as solar energy and geothermal heat. He has also promised to launch an aggressive energy conservation programme.

Carter has also offered a proposal to consolidate most of the departments and agencies now responsible for energy programmes into a single department. Included would be the Energy Research and Development Administration (ERDA)—which Carter has attacked as being overwhelmingly pro-nuclear—the Federal Energy Agency and the Energy Resources Council. The proposal is likely to meet stiff opposition in Congress, however, since many Congressional committees which now have jurisdiction over individual agencies would be wary of losing some of their authority. A prolonged battle should be anticipated.

On nuclear proliferation, Carter has repeatedly called for a worldwide moratorium on the sale of reprocessing and uranium enrichment plants, and for stringent controls on the export of other nuclear technologies. He has also proposed that an international conference be held to discuss alternative energy sources.

Perhaps the most important nuclear policy question confronting Carter, however, will be whether to permit spent nuclear fuel from commercial power plants to be reprocessed, and the plutonium extracted from it to be recycled as reactor fuel. Just five days before the election, Mr Ford announced that his Administration would defer a decision on reprocessing and recycling plutonium at least until the matter has been given more study. Many nuclear critics, including Carter, called the announcement "too little and too late", but Carter's own policies for

recycling have yet to be specifically stated.

Finally, Carter has promised to scale down the effort to develop a liquid metal fast breeder reactor (LMFBR), now the largest single federal energy programme. He has stated that he will reduce the effort to a relatively minor level and, perhaps, seek international participation in it. If Carter decides to

reject plutonium recycling and to scale down the LMFBR programme, those actions would constitute major defeats for the long-term plans of the nuclear industry.

● **Biomedical research.** Virtually nothing has been said during the campaign about the federal government's

Six-state nuclear battle: results

ALTHOUGH the nuclear industry in the United States is not too happy about the fact that last week's election will put into the White House a man who is decidedly cool toward nuclear power, it can at least take considerable comfort from balloting in six states. Voters in Arizona, Colorado, Montana, Ohio, Oregon and Washington soundly rejected propositions which would have placed crippling restrictions on nuclear plants in those states. A similar proposition was defeated in a seventh state, California, last June.

The propositions were rejected so decisively—by margins ranging from 58–42% in Oregon to 71–29% in Colorado—that the results constitute a stunning setback for the anti-nuclear movement in the United States. Spokesmen for the nuclear industry in fact wasted no time in claiming the outcome as a popular endorsement of nuclear energy. Carl Walske, president of the Atomic Industrial Forum, said last week, for example, that nuclear power "has been taken to the Village Square, as Einstein predicted, and has been approved by the American voter".

The defeats were particularly striking since opinion polls conducted only a month before the election indicated that the propositions stood a good chance of being approved in most of the states. In Colorado and Oregon, they were even leading by a margin of two to one. Anti-nuclear groups blame their defeat on the lavish funding poured into the campaigns during the closing stages by industry and labour groups anxious to swing voter sympathies against the propositions. Pro-nuclear forces are reckoned to have outspent their opponents by about 8 to 1, and it is noteworthy that in Montana industry groups put up more than \$80,000 to defeat an anti-nuclear proposition, even though Montana has no nuclear plants and no plans to build any.

The propositions would have forced new nuclear plants to meet three tough requirements before being put into operation. First, there

must be no limit to the damages which could be claimed by victims of a nuclear accident — at present Federal law limits the total liability per accident to \$560 million. Second, there must be an acceptable, proven technology for getting rid of nuclear wastes. And third, key power plant safety systems must be found to work satisfactorily. The last two conditions would have to be met to the satisfaction of at least two-thirds of the members of state legislatures. Those requirements are so tough that if they were passed in any state they would have virtually halted nuclear expansion in that state. Consequently, the industry fought them on the grounds that they would increase energy prices, discourage industrial expansion and increase unemployment.

The crushing defeat of the propositions in all seven states could have important national implications. For one thing it is likely to take the steam out of attempts in Congress to curb the growth of nuclear power. In the past few years, several bills aimed at halting nuclear expansion at least until a variety of conditions are met have been introduced. Though they have never enjoyed much Congressional support, if voters had demonstrated their enthusiasm for such measures by passing some of the propositions, anti-nuclear forces in Congress would have been given considerable impetus.

The outcome of the referenda is also likely to discourage similar attempts to limit nuclear power through direct ballot in the future. In fact, some nuclear spokesmen are already claiming that the crushing defeat of the propositions will lead to a sharp decline in anti-nuclear sentiment in the United States.

Although such claims will almost certainly prove to be exaggerated, the outcome of the referenda has clearly provided the nuclear industry with considerable ammunition to combat President-elect Carter's reservations about expanding the use of nuclear power in the United States.

\$2,000 million biomedical research programme, but again, Carter will be faced with some controversial decisions.

First, however, it should be noted that Carter's election may help reduce one of the chief problems which has plagued the federal biomedical research effort for the past few years. Major tussles between Congress and the Ford and Nixon Administrations over the size of the budget for the Department of Health Education and Welfare, of which NIH is a part, have frequently delayed the release of funds for NIH and thrown planning into confusion. The Carter Administration is likely to work more closely with Congress on such matters, and thus NIH's planning should be made easier.

Among the more touchy issues which Carter must sort out are whether to continue to support the training of biomedical researchers—Nixon and, to a lesser extent, Ford tried to phase out the NIH training programmes over noisy opposition from the scientific community. He will also have to adjudicate the growing controversy over whether the National Cancer Institute should continue to enjoy a privileged

position in NIH. In that regard, it may be recalled that Ford took a tentative step by suggesting that the cancer budget should be held constant this year while support for research on other diseases should be allowed to grow, but Congress increased the cancer budget anyway.

● **Arms control.** Though many of Carter's foreign policy statements have a distinctly hawkish ring about them, he has also criticised some arms control treaties as being inadequate. In particular, he has called for a complete cessation of nuclear testing, and he has labelled the proposed Threshold Test Ban Treaty as "totally inadequate". That treaty is now pending before the Senate for ratification, and it is likely that Carter will try to negotiate something more substantial instead of trying to get the measure through the Senate.

● **Congress.** Several key Senators and Congressmen who have been influential in science policy affairs have either retired or were defeated in the polls last week. The chief casualty was Senator Frank Moss, chairman of the Senate

Committee on Aeronautical and Space Sciences. But perhaps the most important changes are likely to come in the powerful Joint Committee on Atomic Energy, where the chairman, John Pastore, and Stuart Symington have both retired, and Senators Joseph Montoya and John Tunney were both defeated. On the House side, James Symington, the chairman of the subcommittee which oversees the work of the National Science Foundation, relinquished his House seat to seek election to the Senate. He has been an important defender of NSF during the past couple of years, but at least his loss will be partially balanced by the fact that John Conlan, NSF's chief critic, also relinquished his House seat to run unsuccessfully for the Senate.

Though many important science policy issues have been virtually ignored during the election campaign, Carter's handling of them in the next few months will be closely watched. Leaders of the scientific community are not usually reticent in giving policy advice to the government, and they can be expected to make themselves heard in the coming weeks. □

AUSTRIA

Nuclear consultation

Engelbert Broda writes from Vienna on Austria's nuclear energy information campaign.

UNPRECEDENTED action has been taken by the government of the Republic of Austria. In view of the wide divergence of opinions on the desirability or otherwise of nuclear energy and the emotions aroused, it has decided to consult the population on the matter directly. Although decisions will eventually have to be taken by Parliament, the government is making sure that the views of the people can serve as a basis. A nation-wide information and discussion campaign has been organised, financed by the office of the Prime Minister, Bruno Kreisky, in collaboration with the "Energy Section" of the Federal Ministry of Trade and Industry. The section is headed by Wilhelm Frank, a man with wide scientific interests who is not only a professional mechanical engineer but also a renowned theoretical mathematician and a philosopher.

Austria's energy position is relatively favourable. While for many years power consumption increased by about 7% per year, the rate of increase has, as elsewhere, gone down more recently. Hydroelectricity still accounts for more

than 70% of total electric power, and there are still unused reserves of hydropower, mainly along the Danube. (It is interesting, though, that a popular movement for the protection of the Wachau, the part of the Danube valley with the most beautiful scenery, has induced the government to shelve and reconsider plans for the exploitation of that stretch.)

The production of brown coal, mainly in Styria, is considerable and goes mostly to power plants near the pits. While normally some electric power is exported, in emergencies power can also be imported by high voltage transmission lines from west and east. About one-fifth of the oil and gas used in the country comes from wells in Austria. The necessary imports are widely spread geographically: sources include the Middle East and Russia. One nuclear plant, a 700 MW boiling water reactor on the Danube about 40 km north-west of Vienna, is in an advanced stage of construction.

That the first nuclear power station will be taken into exploitation is hardy in doubt. But although detailed projects exist for further stations, no decisions have been taken as yet. At a press conference held soon after the elections in Sweden that were so influenced by the nuclear issue, Austria's



Austria's Chancellor, Bruno Kreisky

Federal Chancellor cheerfully said that he was doubtful about the wisdom of more nuclear power, while the Minister of Trade, Herr Staribacher, expressed himself in favour of additional stations. In such circumstances the voice of ordinary people will clearly have a lot of weight.

The Federal Press Service has issued a little book containing basic information, as objective as possible, on nuclear power. There will be 10 public meetings in various towns where the members of the public may ask questions and offer comment for up to 90 seconds each. Each meeting deals with a different aspect of nuclear energy—

socio-economic problems, energy policy, safety, social repercussions, influence on climate and biomedical problems. The discussion on each subject can be held in one place only, but written contributions are also accepted.

Each meeting is prepared by a subgroup of five or six experts, including natural scientists, sociologists and economists. West Germans, Swiss and even an eminent Bulgarian are among the members. In preparatory meetings, each group tries to reach factual agreement as far as possible but does not hide internal divergences when it faces the meeting. Disagreements are bound to be pronounced as in every case both friends and foes of nuclear energy have been appointed to the groups. They do

not include, however, members of the nuclear establishment, notably employees of the companies that build or plan nuclear power stations.

The first public meeting was extremely lively. Following the meetings each group will submit an extensive report which will be further discussed publicly between adversaries and supporters of nuclear power. No party whip will be applied in Parliament, the Chancellor has said.

If in the end a major expansion of nuclear power production were rejected, Austria would have to take drastic steps to curb the further increase in power, and energy consumption generally. Alternative sources of energy, save undesirable imports, are in

the short term limited though the long term prospects for solar energy seem good. There is no doubt, however, that the scope for energy conservation in Austria is enormous: energy has been anomalously cheap, with oil prices low until 1974 and the sales price of electric power not keeping pace with inflation.

One perhaps inevitable, if regrettable, feature of the information campaign is the near-absence of a crucial issue: the subject of the plutonium economy and weapons proliferation. This problem is not so obvious to the citizens of a small and neutral country that spends little on the military; the success or otherwise of the bold venture they have embarked upon, however, will be watched closely. □

REPROCESSING

Permission to process

Gillian Boucher reports on recent developments in plans for the reprocessing of nuclear fuel in Britain.

THE decision last week by Cumbria County Council's Planning Committee to approve in principle the £600-700 million programme for the expansion of the nuclear fuel reprocessing plant at Windscale brings Britain perceptibly closer to becoming a world reprocessing centre. Unless Mr Shore, the Secretary of State for the Environment, exercises his option to re-examine the question within 21 days of receiving notification of the committee decision, outline planning approval is automatic. That will remove probably the greatest single impediment to the signing of the controversial nuclear reprocessing deal with Japan.

To gain planning permission will be an important victory for the nuclear industry which, since the negotiations with Japan were publicised at the end of last year, has been fighting a noisy battle with well-organised environmental groups. Fears centre on the prospect of Britain taking on long-term storage of other countries' fuels which she has reprocessed—becoming what the popular press has called the world's nuclear "dustbin"—and the dangers of sabotage if plutonium is shipped back to the country of origin.

After a five-month delay to the negotiations with Japan to allow time for public debate, Mr Anthony Wedgwood Benn, the Secretary of State for Energy, announced in March of this year that foreign deals would be acceptable if Britain did not have to store the fuels on a long-term basis but had the option of returning them

to the country of origin. But the Flowers Commission said in its report on nuclear power and the environment that it would be unwise to take up the option, that it was best to avoid transporting the wastes if possible and that long-term storage in geological formations would almost certainly be safer in the UK than in seismically active Japan.

The deal originally being discussed with the Japanese utilities was for British Nuclear Fuels Ltd (BNFL), the state-owned company which operates Windscale, to reprocess 3270 tonnes of spent Japanese oxide fuel during the 1980s. The deal was worth around £4000 million including a large

down-payment. But the delay for public debate gave the French reprocessing organisation, Cogema, a chance to declare its interest; both BNFL and Cogema, a subsidiary of the French Atomic Energy Commission, are partners in United Reprocessors, an Anglo-French-German market-sharing body. The final contract, now likely to be signed in the next few weeks, will probably share the Japanese work equally between Britain and France. British contracts with other countries are also being negotiated.

BNFL has had government approval of its plans to invest £245 million on extending and improving the existing Magnox fuel reprocessing plant and £40 million on development of the vitrification process; it still awaits the crucial approval of the £350 million



Windscale reprocessing plant

investment on Thorp, the oxide fuel plant, which will be vital if Britain is to become a big exporter of its reprocessing services. If business prospects continue to look good, permission will almost certainly be forthcoming.

Cumbria County Council has had a daunting task in assessing the application for planning permission—weighing on the one hand 2,000 jobs in an area of high unemployment and the prospect of much-needed foreign earnings, some of which have already been lost to France through delays, against unfamiliar dangers to a rural community on the other. The Windscale workers have been staunchly in favour of expansion and wanted the County Council to make its decision without further delay. But Friends of the Earth, Half-life, the Conservation Society and the Town and Country Planning Association among others, concerned principally with the oxide plant, wanted the matter referred to a public enquiry.

In the end the committee passed a resolution saying that it was "minded to approve" the application "subject to agreement of appropriate conditions" but passed the final responsibility over to Mr Shore as the application was a "departure from a fundamental provision of the county development plan". At the beginning of this week the Department of the Environment said it had still not received formal notification of the Committee's decision. The pressure groups will continue to call for a public inquiry in the little time remaining.

BNFL has been accused by the Lawyer's Ecology Group of providing

Why reprocess?

THE short answer is to make plutonium for fast breeder reactors. Though fast breeders will make us virtually independent of U_{235} sources, the supplies of plutonium and depleted uranium produced by the present generation of thermal reactors are essential for them. But much depleted uranium is treated before use in thermal reactors. Reprocessing also makes environmental sense. The 30% waste left after the removal of uranium and plutonium is more compact and easily stored than the untreated spent fuel.

A reprocessing works is essentially a straightforward chemical plant with elaborations necessitated by the radioactivity of the materials. The burnt fuel (uranium metal in the case of the Magnox reactors currently in use in Britain, uranium oxide for other reactors including AGRs and PWRs) is dissolved in nitric acid and the uranium and plutonium extracted with an organic solvent. The remaining highly active acid solution containing a variety of α and β emitters is at present stored in elaborate stainless steel tanks.

Incorporating the wastes in a solid would allow less hazardous storage, perhaps in deep boreholes either on land or under the sea. Solidification

also makes long-distance transport feasible and, if transport of wastes back to their country of origin is made compulsory, will be the *sine qua non* for Britain's role as a major international processor of nuclear fuels. A process of vitrification developed at Harwell in the late 1950s and known as Harvest has been taken up by British Nuclear Fuels who hope to be constructing a demonstration plant at Windscale by 1980. But the technological prospect is daunting, combining as it does intense radioactivity with the high temperatures needed for glass making.

Another large question mark hangs over the business of processing oxide fuel which, being more highly irradiated, poses greater problems than Magnox fuel. At present there is no commercial scale oxide fuel reprocessing plant in operation anywhere in the western world, though the La Hague plant opened earlier this year is approaching that level. At Windscale a "head-on" plant for pre-processing oxide fuel before it joined the Magnox fuel was in operation, but an accident in 1973 in which a number of workers were contaminated led to the closure of the plant. It is expected to reopen next year.

the County Council with "the sketchiest ever (application) ever offered for any major development" and of leaving it "until all the options were closed". But although the replies to the County Council's detailed questions were only

just ready in time for the public debate, Professor J. H. Fremlin, the County Council's independent scientific consultant, found he had all the information he needed to assess the dangers to the public at large. □

BRITAIN

A case of suitable treatment?

A number of members of the World Federation of Scientific Workers received inexplicable treatment from the Home Office when they tried to enter Britain for the federation's 11th General Assembly on September 17–23. Professor Eric Burhop of University College London, who is President of the federation, gave Nature his view of what happened

THE Federation's General Assembly, its highest policy-determining body, meets every three years. This year, on the occasion of its 30th anniversary and amidst some difficulty for many participants, the assembly was held in London, where the organisation was founded. Professor Burhop tells his story with the aim of preventing another recurrence in the future—the

assembly had once experienced even more serious difficulties some 15 years ago.

Before the latest meeting, says Burhop, a British MP associated with the affiliated organisation ASTMS explained to the then Home Secretary, Mr Roy Jenkins, the nature of the planned meeting and the countries from which the attending scientists would come. That was in February. Mr Jenkins' reply, according to Burhop, stated that he could see no reason why the assembly should not meet in London; but he added that people coming from countries from which visas were required should make their application in good time and certainly not less than two weeks before they wished to enter Britain, to give time for the necessary processing. "We informed our affiliated organisations

accordingly," says Burhop, "and expected there would be no more trouble. It was not until September 14, three days before the Assembly was due to commence, that we began to receive cables from delegates from distant parts that their visas had not been issued."

Burhop is quick to repudiate the frequent charge that the federation is a communist-controlled or Soviet-dominated organisation. That is not true, he says, and never has been. He summarises the aims of the federation as international scientific cooperation, the protection of the freedom and rights of scientific workers, the constructive application of science and international agreement on disarmament and the eventual abolition of nuclear weapons. He describes the attempts of the federation after the Second World War to hold meetings in London to discuss nuclear weapons as "sufficient to send sections of our

military and political establishment into a flat spin”.

Official attitudes appear to have relaxed in recent years, but the federation's latest troubles, as told by Burhop, suggest that it is still viewed with deep suspicion. First to complain of difficulties was an Egyptian professor who had applied in Cairo on July 25 for his visa. Burhop says he received the last of several telegrams from the professor on September 16, saying that owing to the non-arrival of his visa he had cancelled his flight. The professor's visa was finally granted three hours before the commencement of the first meeting he was due to attend. A professor from Vietnam was the next in difficulties. He had applied for his visa five weeks previously at Hanoi, and it was granted on September 16—too late to catch the weekly direct flight from Hanoi to London.

Both professors had visited Britain previously without any difficulty, the Egyptian on many occasions. They were each able to reach London by circuitous routes, but some two days late. Four scientists who formed the delegation of the Organisation of Korean scientists in Japan were not so fortunate. They were members of the community of North Koreans who were stranded in Japan after the Second World War and not allowed to return to North Korea; unlike Japanese nationals, they needed visas to enter the UK, and re-entry permits to return to Japan. The Home Office gave the date of receipt of their visa application as September 2, which allowed the required two weeks for processing; the scientists claim that they applied even earlier. Their visas were granted on September 22, the day before the assembly ended.

But as Burhop points out, Home Office procedures were shown in the oddest light in the handling of the Soviet applications. After the French delegation, the Soviet delegation was the second largest at the Assembly. It consisted of twelve persons, including two Academicians, one a Nobel laureate and another a famous mechanical engineer, a James Watt medalist of the British Institution of Mechanical Engineers. Both members of the Supreme Soviet of the USSR (one is on its Presidium), they had visited Britain many times.

Burhop takes up the story: “There is a discrepancy in dates when the applications are said to have been made. Our Soviet colleagues claim that their Ministry of Foreign Affairs transmitted all the applications together to our Embassy in Moscow on September 3, exactly two weeks before the date of travel as specified by Mr Jenkins. The Home Office claims that they were received by our Embassy only on

September 8. Late on September 17 I was informed by the Home Office that three of the Soviet visas had been granted, the rest were under consideration, and no more would be granted until Monday. Of the three visas granted two were of secretaries to the delegation. The other was a social scientist and authority on disarmament. The Soviet delegation that sets great store on its distinguished Academicians was deeply offended. It read into the decision a studied insult, which I do not believe was intended, and I feared at one time it might develop into a diplomatic incident. The remaining Soviet visas, with one exception, were granted about midday on September 20, in time to permit the whole delegation with the exception of the scientist whose visa was refused to travel to London that evening. They were able to make an effective contribution to the rest of the Assembly.”

Then there was the GDR delegation, which included the President of the GDR Academy of Sciences and a university rector. Their application was late, and was received only on September 10. The Polish delegation, however, applied at the same time and received visas almost on time. Visas for the whole of the GDR delegation were granted only on September 21. They arrived the following day and participated only in the tail-end of the assembly.

Burhop again takes up the story: “Of course I do not excuse those delegations who applied late. But some of those who applied very early were also very late in receiving their visas. I was informed by the person handling the visas at the Immigration Department that all the visa applications had arrived on his desk only on September 15 and only then could they begin the usual processing. This was despite the fact that as early as July our office in London had started supplying to the Home Office lists of all the delegates coming from abroad, details concerning whether they needed visas or not and, by early September, the name and country of each participant.”

What went wrong? The change of Home Secretary on September 15 may have caused difficulties. The new Home Secretary, Mr Merlyn Rees, was absent from London on September 17 and September 20; it may be that the civil servants whose responsibility it was to take the decision were not willing to do so in the absence of the new minister. But Professor Burhop feels that no matter when the visas were applied for it made no difference: processing of them only started two days before the assembly was due to start and proceeded at a leisurely pace.

The most curious case of all is that of the Soviet delegate, Professor

Grigori Kotowski, whose visa was refused. One newspaper telephoned the Home Office and was told that Professor Kotowski was only an organiser, not a scientist at all. But according to Professor Burhop and others he is a social scientist of great eminence, deputy head of the Institute of the Peoples of Asia of the USSR Academy of Science and a leading authority on India. Kotowski has frequently visited the UK without any trouble, only last year attending on behalf of the USSR an executive meeting of the International Economic History Association.

Burhop speculates that the refusal of his visa was conceivably a matter of bureaucratic face-saving. Having boomed about the issue of visas the Home Office felt it had to refuse one to give the impression that there were very serious reasons for the delay. Alternatively—and less flattering to the Home Office—Burhop suggests a mix-up. “The name Grigori Kotowski is very well known in the Soviet Union. It is that of an almost legendary hero, a partisan leader, famed for his military exploits in Moldavia during the Civil War. There is an imposing statue to him in Kishinev. A tank regiment in the Red Army is named after him.

“Professor Grigori Kotowski is his son. A less military looking man or a man more academic or professorial in bearing than Professor Kotowski it would be impossible to imagine. But dare I suggest that the Home Office have got the records mixed up and that when they refused a visa to Professor Grigori Kotowski they mistook him for his father? Of course there is a small matter of fifty years between the careers of the two Kotowskis. Nevertheless in all this matter the Home Office has seemed so maladroit that nothing would surprise me.”

The Home Office itself, when contacted by *Nature*, was hardly illuminating. The applications from North Korea, it admitted, did take a “little bit longer” than others. “We’re sorry about it”, a spokesman said, but “it took this time to clear. We take our responsibilities very seriously”. Asked about the case of Kotowski, he explained that reasons were not given for the refusal of a visa; it depended a lot “on the relationships between one country and another”. But he made a suggestion: why didn’t *Nature* give the number of people who did receive their visas on time?

The response is printable. Of the 100 or so people attending, 41 required visas to enter Britain: 16 received their visas in time, 20 received them late and so missed at least 2 days of the assembly, 4 received visas too late even to attend, and one had his visa refused.

□

SCANDINAVIA

Controlling the pollution trade

Under a unique treaty which entered into force last month, private citizens living in the Nordic area will have the right to complain about and be compensated for damage to their environment caused by activities in their neighbouring countries. Mike Duckenfield reports

THAT the first agreement of its type should be reached between the Nordic nations is no surprise. Norway, in particular, suffers badly in the present European exchange of pollution and the Scandinavians as a whole are keen to secure international consensus on its control. In deciding to put their own house in order they are not only showing their seriousness, but aiming to pre-empt any criticisms about their own lack of action.

The agreement, prompted by the Nordic Council and covering all five of its members except Iceland, will be supplemented next January by Norwegian legislation requiring industry to use low-sulphur fuels. In October next year a start is due to be made to a seven-year Swedish plan to cut sulphur emissions to their level in the early 1950s.

The Swedish plan follows a recent government commission appointed by the Minister of Agriculture which carried out an extensive survey of air pollution problems caused by the burning of fossil fuels containing sulphur. It estimated that in 1973 about 60 million tons of sulphuric fallout, either dry or in precipitation, was generated in Europe, compared with only 25 million tons between 1910 and 1950. Of this, 38 million tons came from Eastern Europe and the remainder from the West. In addition, increased energy consumption in all countries was expected to result in the generation of 74 million tons by 1985, with 27 million tons coming from the West.

In 1974, Sweden itself burned about 19 million tons of fuel oil, two-thirds of it heavy. As oil with a sulphur content of 2.5% by weight causes emissions of about 50 kg of sulphur for every ton of oil burned, it was calculated that about 800,000 tons of sulphur was emitted, a quarter of it from industrial processes using sulphuric compounds and the remainder from burning oil. If no action was taken this would rise to one million tons by 1985. As it is, the aim is to cut it to 300,000 tons.

To achieve this the Swedish plan involves banning the import and sale of light fuel with more than 0.5% sulphur

nationally from next October and, at the same time, prohibiting the use of heavy oil with more than 1% sulphur in the part of the country south of a line north-west of Stockholm. Two years later, the ban would extend to light oils with more than 0.3% sulphur and, in 1981, the extension nationally of the heavy oil ban. The plan also requires that the existing ban on burning oil with more than 2.5% sulphur be placed on other fossil fuels, most notably coal.

The programme would cost an estimated 500 million Swedish crowns (about £72 million) and includes government grants to finance the experimental liming of some of the 10,000 or so lakes that have been acidified by sulphuric fallout. At present it costs between Skr 400 and Skr 500 per hectare to lime the lakes and the job has to be repeated every five years.

For all this, Sweden expects to be able to do no more than maintain the *status quo* in the worst affected southern and western parts of the country. The reason is simply that most of the pollution is imported, mostly from the south and west across the North Sea and Baltic. In these areas it is estimated that less than 25 per cent of the fallout is Swedish in origin. In the remainder of the country the figure varies from between a quarter to a half of the total. Nationally it is just short of one-third. Because of this, the Swedes would now like to see total emissions in Western Europe brought down to between six and eight million tons within the next 10 years, a task that would cost about Skr 20,000 million a year.

In neighbouring Norway the situation is even worse with less than one-tenth of fallout being home-produced. Even in the area around Oslo, the country's most industrialised region, the figure only reaches 30-40%. The thin soil over much of the country does little to neutralise the acid rain and the build up of polluted winter snow leads to an annual mass slaughter of thousands of hatching and spawning fish when the spring thaw pours into the rivers and lakes. In the worst-hit south-west salmon and trout have disappeared from many rivers and a recent survey of 153 previously well-stocked lakes revealed the eradication of all types of fish from all but two of them.

So serious has the problem become that the Norwegian government is currently spending about £6.5 million in employing 60 full-time scientists on a seven-year programme to examine the effects of acid precipitation. And to draw international attention to the

need for control, the Norwegian Ministry for the Environment together with the precipitation project (SNSF project) this summer invited representatives from 20 nations including all but Luxembourg of the Common Market countries, the Soviet Union and three Eastern European nations to a conference at Gaustadblakk near Rjukan in Telemark in the south-west.

Research shows that not only are fish being killed, but acid rain is affecting soil productivity and could be causing decreased forest production due to the depletion of the earth's nutrients. Similarly, the Swedish commission report also points to the effects on urban areas due to the corrosion of metals and stone, especially limestone and marble. Costs of quicker replacement of materials and the need for corrosion protection were put at anywhere between Skr 100 million and Skr 1,000 million annually, not including the irreplaceable damage done to historic buildings and monuments.

While the conference could only agree to recommend nations to reconsider their approaches to emission control and to sponsor more research, international cooperation may be given a much needed boost with the publication of a report by the Organisation for Economic Co-operation and Development next year. OECD's recently completed research project into the long-range transport of air pollutants will show just how badly affected southern Scandinavia is. In one extreme case in January 1974, winds from all over Europe brought 4,000 tons of sulphate and dumped it over about 20,000 square km of southern Norway in just 12 days. Some areas had nearly a ton of sulphate per square kilometre. □

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Pollution demo, Swedish-style

Keystone

IN BRIEF

Appointment of GMAG chairman

The first chairman of the British Genetic Manipulation Advisory Group is to be Sir Gordon Wolstenholme, Director and Secretary to the Trustees of the Ciba Foundation and President of the Royal Society of Medicine. The establishment of a group to advise laboratories on appropriate precautions in genetic experiments was the main recommendation of the Williams working party which reported in August this year. The group will examine proposals from laboratories (though precisely which types of experiment will have to be submitted is yet to be decided), assess the hazards involved and advise on appropriate precautions. The names of other members of the group will be announced soon.

Customers for nuclear club?

The secret club of 13 nuclear exporters, which aims to restrict nuclear exports to countries which have not ratified the 1970 Non-Proliferation Treaty, is reported to be considering admitting customer nations to its circle. The group is expected to meet in London this week. Customer nations made their resentment of the group's clandestine activities clear at the International Atomic Energy Agency annual conference in September; it has been suggested previously that the group admit customer nations but some members, notably France, were against it. Iran is being suggested as a possible first customer-member, as its large nuclear programme involves exports from several countries including the United States, Germany, France and the United Kingdom.

French chemical controls

Production and import of chemicals in France will be much more strictly controlled in future if a draft law adopted by the Council of Ministers last week is passed in Parliament. Producers and importers will be required to make a study of the effects of new chemicals on man and the environment and make their results available to the authorities, who will have six months to decide whether the manufacture, sale or use of the chemical should be subject to regulations or even banned completely.

The measure will fill a gap in existing legislation, which regulates the use of specific products such as explosives, pesticides and food additives and protects certain defined groups. The new law will be the first of its kind in Europe.

BRAZIL is a country of extremes; wealth and poverty, grandeur and squalor, but above all, tremendous vitality and optimism. What other country could build such a fantastic new capital as Brasilia? Brazil is completely fascinating to a biologist, whether standing on a street corner in downtown Sao Paulo and watching the complete ethnic diversity of the hurrying crowds, or gazing from a plane at the unbroken green carpet of wild vegetation stretching for hundreds of miles below, without a sign of human impact. Or seeing the "mud house" nests of a charming bird, Joao de Barro, on the crossbars of most of the power line poles along the highway going south from Porto Alegre.

Brazil is where the high technology of the 1970s is brought to bear on one of the last great untouched areas of the earth. Can we hope that enlightenment will come in time to enable this country to escape the rapacity of human beings and their new machines? Sometimes, the results of change are obviously beneficial. Large numbers of Japanese immigrants have come to Brazil in recent years. The farmers among them were accustomed to producing large yields from their tiny farms in Japan. They found that land was plentiful in Brazil, and they evidently apply the intensive methods they used in Japan to much larger farms. (Brazil is the world's fourth largest market for pesticides.) The result is that the huge city of Sao Paulo now receives a plentiful supply of splendid fruits and vegetables, probably for the first time.

I saw these commodities exchange-

ing hands in the big wholesale markets at midnight. The produce was as carefully packed and graded as if it had been intended for gift shops. The nutritional status of many Brazilians must have been greatly improved by the industry of these new farmers. But in some regions, malnutrition is

Brazilian dilemma



THOMAS H. JUKES

common. When I visited the Ministry of Health in Brasilia, I was told that vitamin A deficiency is so prevalent in north-eastern Brazil that the changes produced by it obscure the "Pap test" in women. Also, the deficiency causes blindness in many children. For people who cannot or will not eat green leaves, synthetic vitamin A should be added to food. It costs about three cents per million units—about \$150 for one million adult Recommended Daily Allow-

ances or for two million for children under four.

The huge Volkswagen factory at the western edge of Sao Paulo has helped to produce an "instant middle class" in Brazil. On city streets, drivers blow their horns and flash their lights at each other, to challenge the right of way, except when, as often happens, traffic is completely clogged.

A great question for the future of the biosphere concerns the fate of Amazonia, which includes six states and portions of three more. The Companhia Industrial de Amazonia was formed in 1966 for producing non-ferrous metals; Sudam is the agency for development of Amazonia including plans for iron ore and bauxite, for hydroelectric power, timber-cutting and raising cattle.

Twenty per cent. of the world's fresh water supply flows through the Amazon basin network of rivers. Amazonia's area is nine times that of France. Its colossal jungles have a canopy of green vegetation above a thin layer of fragile soil and resting on hardpan. When the jungle is cut down the topsoil tends to disappear, leaving an infertile base. It seems that little is gained by destroying the jungle, and at stake is one of the world's great photosynthetic factories for maintaining atmospheric oxygen. The birth-rate of Brazil, where Roman Catholicism is the state religion and divorce is not permitted, results in a population growth of about 2.8% annually.

Will a desire to preserve Amazonia override the pressures for Brazil's western expansion?

correspondence

Biological fly killer

SIR,—K. M. S. Aziz (October 14, page 544) misquotes F. E. G. Cox (August 19, page 646) when referring to the use of insecticides for the eradication of tsetse flies (principally *Glossina morsitans*) from some 27,500 km² of riverine vegetation in Nigeria. This was achieved by ground spray techniques and not by aerial spraying.

This is a point of detail and Aziz could well reply that it does not affect his argument that aerial spraying (as practised against *Glossina* elsewhere in Nigeria and in other African countries) is an unsatisfactory method of controlling insect species. He gives as an example the resurgence of mosquitoes after an aerial spray operation over parts of Bangladesh. However, mosquitoes in Bangladesh present different problems to tsetse flies in Nigeria and it can be misleading to compare the two situations.

I suspect that the mosquitoes of Bangladesh re-appeared because the aerial spraying operations had no permanent effect on their habitats. Precisely the same would almost certainly have happened in time in the case of the tsetse flies of Nigeria had the habitat remained unaltered. However, Nigeria is a land-hungry country and the success of the anti-tsetse operations allowed a breathing space in which land cleared of *Glossina* was taken over by man and his cattle, altering the vegetation to such an extent that it no longer presented a suitable habitat for *Glossina*.

The moral of the Nigerian success story is that tsetse control operations should not be initiated unless there is a demand for the land occupied by the flies. Obviously, in order to prevent such evils as overstocking and soil erosion, exploitation of cleared areas must be strictly governed by well thought out development programmes.

Insecticides for tsetse control do have, and will continue to have for the foreseeable future, an important part to play in the development of rural Africa. By all means let us undertake research, as advocated by Aziz, on more specific biological methods of control; but Africa cannot afford to postpone action against *Glossina* until satisfactory alternatives to insecticides have been developed.

Your faithfully,

A. M. JORDAN

Tsetse Research Laboratory,
Langford, Bristol, UK

Nobel prizes

SIR,—I have found what I believe to be a significant correlation between Nobel prizes and Olympic games. Previously, the United States did not have the dominant position in science that it has achieved today. The Nobel prizes of physics, chemistry and medicine were never awarded to US scientists in the same year; now the situation has changed. In 1968 all three prizes were for the first time awarded to American scientists. The second favourable conjunction came in 1972 (seven titles out of eight, in the three domains). Now in 1976 there is again an American monopoly. The three remarkable events are significantly correlated in time with the Olympic Games.

Some friends of mine suggest that the

truly significant correlation is to be made with the presidential elections in the United States. Such a hypothesis is easily ruled out on the basis of the authoritative works of Einstein and Garfield. It is known that the presidential elections occur after, not before, the designation of the Nobel laureates. Einstein's theory of relativity (*Ann. d. Phys.*, 17, 891; 1905) excludes any time-reversed determinism. Then, the objectivity and political independence of Stockholm's academicians is attested by the fact that the Nobel prize correlates very well with the value of the distinguished scientific work, as measured in the Science Citation Index (Garfield, *Nature*, 242, 485; 1973).

Yours faithfully,

JACQUES NINIO

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Stamps of scientific interest

On October 9 Sweden issued a colourful set of five stamps commemorating nineteenth century Swedish pioneers in various spheres of technology. Each of the stamps bears a portrait of the pioneer in question together with an example of the practical application of his contribution to technology.

These range from work on the use of solar energy (John Ericsson), a machine gun and hay-mower (Helge Palmcrantz), the first desk telephone (Lars Magnus Ericsson), a milk separator (Gustav de Laval) to the first spherical, self-regulating ball bearing (Sven Wingquist). It was this latter innovation which was later produced by the Swedish Ball Bearing Company (SKF), in Gothenburg, which was founded in 1907. L. M. Ericsson's first desk telephone of 1877 was the start of the L. M. Ericsson Telephone Company, now a world-wide enterprise.

Also issued by Sweden on October 9 were two stamps devoted to industrial safety. The first law on this subject in Sweden was passed in 1889, it also being then that the first inspectors were appointed. The present body both investigates hazards on the factory floor and carries out research on occupational health. The design of these two stamps consists

of two hands against a background of two cog wheels. The fingers and the cogs join to symbolize the interplay between man and machine.

Ian F. Finlay



news and views

Joint Oceanographic Assembly

from C. M. Yonge

The 4th Joint Oceanographic Assembly was held in Edinburgh on September 13–24. Thirteen organisations participated under the auspices of a group of international bodies led by the Scientific Committee on Oceanic Research of the International Council of Scientific Unions.

WHAT immediately emerges from these meetings is the enormous recent extension of knowledge in effectively all aspects of marine science. This comes very forcibly to one who worked at the Plymouth Laboratory when W. R. G. Atkins and H. W. Harvey were elaborating the first colorimetric methods for the quantitative determination of phosphates and nitrates which led to elucidation of the annual cycle of events in surface waters, and who first visited the now mighty Scripps Institution when this consisted of a single laboratory, pier and a few scattered staff houses!

Geology which was then of minor concern has since assumed supreme importance. The first impact of plate tectonics is now past. With the accompanying study of sediments, now cored in supreme depths by the *Glomar Challenger*, aided by isotope detection and carbon dating, it provides evidence for the history of the oceans which was inconceivable a generation ago.

The oceans have long been divisible into great latitudinal zones with surface cyclonic or anticyclonic circulations and underlying compensating currents, the deepest waters creeping slowly in an equatorial direction. Upwelling off continents and between diverging current systems appeared as the major means of surface enrichment. W. H. Munk (Scripps Institution, San Diego) now reveals that the greater part of oceanic movement is highly variable, that an a.c. rather than a d.c. circulation prevails. A series of casual currents produces eddies in midwaters. In the Atlantic, some appear to originate by the cutting off of marginal areas of the Gulf Stream but in

general their origins are obscure. Of their great significance there seems no question, absorbing, as Munk calculates, more than 90% of oceanic energy. The superficial zone of high productivity is revealed by infrared photographs taken from satellites. All previous estimates of marine productivity may need considerable modification.

Wartime studies associated with submarine detection revealed the ubiquity and variety of submarine noises, readily detectable because sound pressure in water is a thousand times greater than in air. As L. M. Brekhovskikh (Academy of Sciences of the USSR) reminded us, while we, on the surface, cannot hear the fish below, anything we say may be carried to them. Internal probing of the sea by high frequency ultrasound can provide an acoustic image where no optical image is possible. Meanwhile a widening series of surface measurements—of temperature, currents, wave heights, chlorophyll and sediment concentrations and of much else—become possible from the orbiting platforms of satellites.

The complexity of the marine environment and so of the biological processes within it was evident in many contributions. The greatest of fisheries is that for the anchovetta in the rich and cold upwelling waters off Peru. Productivity is irregularly affected by incursions of warmer waters, the dreaded 'El Niño', from the north. But what had appeared as a direct effect on the fish—and so on their original exploiters, the guano-producing boobies, cormorants and pelicans—now appears as much more complex. The actual nature of 'El Niño' is in question as reported by G. L. Kesteven and L. J. V. Gonzales (Instituto del Mar, Lima). The effect may be less on the adult fish than on their reproduction and on development and survival of young stages possibly predated by medusae and pelagic crabs. Already it seems that the relatively very low spawning stock of anchovies in 1975 has produced more abundant progeny than the more

numerous stocks in the sixties. Until the precise nature of controlling processes becomes clear there can be no certainty about future stocks.

The long term variability of conditions in the Baltic, as indicated by the known vagaries in the herring fishery since the Middle Ages, now fits within a more recently traced pattern of ocean change in the North Atlantic. Warming of these waters with accompanying northward extension of major stocks of food fishes, including development of a cod fishery off the west coast of Greenland, has been followed by recent cooling and withdrawal.

A discussion of conditions in the intensively studied North Sea by A. Lee (Fisheries Laboratory, Lowestoft) provided some insight into what can happen when a body of water is bordered by areas of intense industrial activity. Here the water is exploited for fish, the bottom deposits are a source of aggregates for use in the building industry and the deeper deposits contain large quantities of oil and gas. On the credit side, although domestic and industrial effluents enter in vast quantities, extensive testing has failed to indicate any serious accumulation of pollutants in coastal animals. In the North Sea the major risk comes from escape of oil, directly from the borehead or by fracture of a pipe line. In the open oceans there is a possible future risk when pollutants slowly building up in the profound depths regain the surface.

Removal of wide areas of gravel could certainly have devastating effects on bottom-living animals and especially on the deposited eggs of herrings. The closure of shallow water areas such as the Wash and the Dutch and German Waddensee—both possible—would destroy the nursery grounds of plaice and other food fishes.

The widely publicised destruction of herring stocks, due largely to their over-exploitation for industrial purposes, has strangely been accompanied by great increase in the stocks of mid-water and bottom-living round fishes, such as haddock, which also spawn a

year sooner than formerly, making nonsense of earlier predictions of future stocks. The phosphate content in the southern North Sea has actually doubled in the past 15 years. Here we encounter what must surely be a positive result of human interference. Opinions vary between explaining this as the result of increasing discharge of largely domestic effluents from the surrounding countries or as an indirect consequence of the destruction of the herring fisheries by way of a now much better nourished and more numerous bottom fauna.

These are but few of the topics that claimed attention in a programme which ranged from mathematical treatment of data concerning water movements to stratigraphy, and from man's use of oceanic lagoons to the possibilities of ocean engineering and of extracting energy from salinity differences.

One cannot but be impressed by the obviously increasing integration of marine studies which was reflected in the attitude of the participating scientists. Any venture on or in the high seas calls for international plan-

ning and the cooperation of research vessels from different nations, the greatest such achievement to date being the International Indian Ocean Expedition. It follows that assemblies such as this one are essential if scattered workers are to meet and plan common activities.

Yet this assembly met with a sense of impending difficulties or worse. The Law of the Sea Conference falters on but with the eventual possibility that the present freedom to conduct oceanic research may become hopelessly entangled with far extended and jealously guarded international boundaries. As the mineral resources of the land diminish, those of the sea, notably of the manganese nodules—with significant additions of other metals—so abundant in oceanic depths, become increasingly attractive. Here is an extreme case of "haves" and "have-nots" with only a handful of the wealthiest nations able to afford the equipment needed for oceanic research and so obtain information that could be withheld from others. Such problems may well come to preoccupy future assemblies. □

Anything new about reverse transcriptase?

from Karin Mölling

YES, indeed, but the news is somewhat embarrassing! Since the discovery of viral reverse transcriptase in 1970 the problem has been to transcribe native RNA templates into long representative DNA copies. The DNA synthesised has always been small compared with the viral RNA template and, furthermore, a small portion of the genome is represented in a greatly disproportionate amount. Last year some laboratories seemed to have come up with a solution to this problem: the reverse transcriptase under the then routinely applied enzyme assay conditions was starved of triphosphates (Collett and Faras, *J. Virol.*, **16**, 1220; 1975; Rothenberg and Baltimore, *J. Virol.*, **17**, 168; 1976). All one had to do, apparently, was to raise the triphosphate concentration, which resulted in a higher yield of long complementary DNA—admittedly, still in a very low absolute yield. I was probably not the only one who was told, when this news was brought home from a tumour virus meeting, that experiments under the new conditions were prohibited because of the expense of the isotopes! There seemed to be a way round when it was later published that a carefully optimised detergent concentration for disruption of virus would also result in a better

yield of long complementary DNA (Junghans, Duesberg, and Knight, *Proc. natn. Acad. Sci. U.S.A.*, **72**, 4895; 1975). For a year radiochemical companies must have earned a fortune from deoxyribonucleoside triphosphates, as complementary DNA was being synthesised in the presence of high triphosphate concentration in many laboratories with the aim of obtaining restriction endonuclease maps of the viral DNA and of making more representative DNA probes for hybridisation experiments.

Now it becomes embarrassing: recently it turned out, that the problems were not caused by too low a triphosphate concentration but too high a magnesium concentration (Rothenberg and Baltimore, *Cold Spring Harbor Tumor Virus Meeting*, 1976). Magnesium chelates the triphosphates so that they cannot be used by the reverse transcriptase—a problem which can certainly be overcome by increasing the triphosphate concentration. If the observation turns out to be true, lowering the magnesium concentration will obviously do equally well. It may be a consolation for all those researchers who are tempted to consider themselves stupid for not having found this answer themselves that it had to be a laboratory like Baltimore's to come up

with it. However, the prevention of precipitation of triphosphates is not the only effect of magnesium. That "Mg is RNase"—a slogan known in many laboratories—may also be involved. Degradation of the template certainly presents an important problem in its transcription. This idea is supported by another recent study from Spiegelman's laboratory, which shows that addition of pyrophosphate—which probably inhibits RNase—to the reaction mixture gives a high yield of DNA complementary to poliovirus RNA used as a template (Kacian and Myers, *Proc. natn. Acad. Sci. U.S.A.*, **73**, 2191; 1976). This result will prompt all those who use reverse transcriptase as a tool for synthesising complementary DNA for hybridisation studies to apply these simple conditions immediately. The product obtained however, is single-stranded DNA; why no double-stranded DNA is synthesised remains unclear. This poses problems for subsequent analysis with restriction endonucleases which predominantly recognise double-stranded DNA.

Why was this work performed with poliovirus RNA? Analogous experiments with genomes from RNA tumour viruses are already in progress. Do they present greater difficulties than those described for poliovirus RNA—which is about the same size as a subunit from an RNA tumour virus (10,000 nucleotides)? The genome of RNA tumour viruses is known to contain obstacles for an enzyme which tries to read its way through completely. It runs into stop signs probably caused by the secondary structure of the RNA. This has recently been reported by Haseltine and coworkers from Baltimore's group who classified the DNA into discrete size groups (Haseltine, Kleid, Panet, Rothenberg and Baltimore, *J. Molec. Biol.*, **105**, 107; 1976). The DNA chains of discrete lengths are all initiated with the tRNA primer at a unique site along the 35S genome and grow by linear extension. The predominant size of the DNA is about 100 nucleotides. Surprisingly, even the longest DNA pieces obtained, (about 3,000 nucleotides) do not contain poly(A) transcripts from the 3' end of the genome. If one tries to compare these biochemical data with the beautiful electron microscopic pictures of the viral RNA which have become available from Davidson's group (Bender and Davidson, *Cell*, **7**, 595; 1976), no correlation between the lengths of the DNA size classes and the structure of the RNA can be detected. The complicated secondary structure of the RNA genome remains at present a specific problem for synthesis of a DNA provirus *in vitro*.

Another problem involved in

reverse transcription of the RNA tumour virus genome is caused by the unexpected position of the RNA primer. Why is the tRNA primer located close to the 5' end of the genome if the polymerase is supposed to start there for reverse transcription of the whole genome (Taylor and Illmensee, *J. Virol.*, **16**, 553; 1975)? There remains only a short way to go for the enzyme (from 3' to 5') until it hits the end of the template. The unexplained location of the primer immediately raised the suspicion that this was the reason for the small size and the repetitive nature of the DNA synthesised *in vitro*. It has recently been shown clearly that in fact the region between the primer and the 5' end of the genome was preferentially copied (Cashion, Joho, Planitz, Billeter and Weissmann, *Nature* **262**, 186; 1976). When fingerprinting the RNA protected by the DNA product they were able to identify the region of the RNA by means of the "cap" which is specific for the 5' end. A series of models have popped up immediately which postulate mechanisms by which the reverse transcriptase could still manage to make a complete DNA copy of the genome. A sequence complementary to the initially synthesised DNA piece may be present at the 3' end of the RNA genome which would allow circularisation of the RNA or bridging between two RNA subunits and continuation of the reverse transcription towards a complete copy. The first evidence for such a model was presented by Haseltine during the European Tumor Virus Meeting in October in Switzerland. His earlier observation mentioned above, that the polymerase jumps to the 5' end of the genome without copying the poly(A) region, gives indirect support to his newest result. A highly conserved constant region on the RNA genome next to the poly(A) tract at the 3' end which had been found in Weissmann's and Duesberg's laboratories while mapping the genome, could be a candidate for the complementary sequence. The chance of matching sticky ends in a test tube would then determine the yield of long DNA transcripts and may be low. This could be another reason why complete reverse transcription of viral RNA *in vitro* is difficult to achieve.

However, one should be able to circumvent this difficulty by applying a simple trick. Controlled heating of the viral RNA will knock off the tRNA primer and a new primer (oligo(dT)) could be bound to the poly(A) stretch at the 3' end of the genome. Why does the reverse transcriptase not synthesise a complete DNA provirus now? Other factors are possibly required? Unwind-

ing proteins involved in DNA synthesis have been well-known in bacteria for a long time and have recently been described in mammalian systems (Herrick and Alberts, *J. biol. Chem.*, **251**, 2124; 1976). Such proteins may be required for extending the RNA template or the complementary DNA for double-stranded DNA synthesis. Recently, the stimulating effect of some DNA binding proteins from chicken cells on the avian viral reverse transcriptase has been reported (Hung and Lee, *Nature*, **259**, 499; 1976). A protein factor associated with plasma membrane preparations from chick embryo cells also stimulates the avian reverse transcriptase (Padhy *et al.*, *Nature*, **262**, 802; 1976). A virus structural protein may even be a candidate for a factor involved in reverse transcription. □

Fire on heathland

from Peter D. Moore

A SECONDARY effect of the hot, dry summer in Britain has been the incidence of large numbers of fires, particularly on heathland and moorland areas. Some areas affected are of high biological interest and value, and conservationists must be asking themselves what the long term effects of these fires will be upon our flora and fauna, and also whether anything could have been done to prevent them.

Especially serious are those fires which have affected our National Nature Reserves, many of which are managed by the Nature Conservancy Council (NCC). These areas have been selected for their diverse biota and are usually the best remaining examples of particular habitats in Britain. Therefore damage to these areas is a matter of great concern. One of the worst examples of such damage during the summer occurred on the Hartland Moor National Nature Reserve in Dorset. This 637 acre reserve near Wareham is largely occupied by dry, *Calluna* dominated heathland. Several rare and diminishing species are found on the reserve, such as the Dorset heath (*Erica ciliaris*) and two reptiles, the sand lizard (*Lacerta agilis*) and the smooth snake (*Coronella austriaca*). As a result of the fire the original population of about 800 sand lizards has been reduced to an estimated 30. The destruction of the habitat has meant that the remaining animals have little chance of survival in the wild, so they have been captured and are being kept in vivaria at the University of Southampton until it is possible to reintroduce them. Even fewer smooth snakes have survived.

This fire occurred despite rigorous precautions. A ploughed, perimeter firebreak isolated the reserve from surrounding areas and there are internal, mown firebreaks. Unfortunately, a public right of way crosses the reserve and the fire began inside the perimeter. The internal breaks proved inadequate as a result of the drought.

Heathland is a habitat which is basically maintained by fire, so its recovery will undoubtedly proceed as in the case of past fires. There are three features, however, which make this fire particularly unfortunate. First, the extent of the burn; about 450 acres (over two thirds of the reserve) have been destroyed. This may slow down the processes of reinvasion and means that the refuge areas are small. Second, the involvement of reptiles which already have fragmented distributions. The heathlands of Dorset are now reduced to about nine extensive tracts and reinvasion from one area to another by lizards and snakes is unlikely because of the barriers of unsuitable land separating the heaths. A third unfortunate feature is the intensity of the fire, which has a serious effect upon the peat areas, such as the valley mires and flushes on the reserve.

Peatlands are particularly susceptible to fires during droughts, and several important British peatland sites have been damaged this summer. Parts of Thursley Common in Surrey are an example, also the raised bog of Glasson Moss in Cumbria. In a raised bog the growth of peat-forming plants and the subsequent deposition of their litter can raise a dome of peat four or five metres above their surroundings. The impedance of drainage by the peat mass results in a raised water table within the dome. Glasson Moss is a good example of such a mire. Part of it is managed by the NCC as a National Nature Reserve and this part (about 140 acres out of a total 600) has been very badly damaged by fire during the summer. Its vulnerability is obvious, being surrounded by old, partially drained peatlands and agricultural land.

Glasson Moss was surveyed for the NCC by Grieg (unpublished report to NCC: 1975) and, although he found that the central areas of the dome were of high quality botanically, he considered that this quality had deteriorated in recent years. It is believed, for example, that peripheral drainage is lowering water tables in the central area and it is known that the surface was badly burned in 1969. The whole *Sphagnum pulchrum* / *Rhynchospora alba* / *Drosera anglica* assemblage, typical of the central dome, is now becoming very scarce on Glasson Moss. Glasson Moss is not alone. Borth Bog in the Dyfi Estuary National Nature

Reserve, West Wales, has been burned again this year and this has become almost an annual event. Many such fires are known to be started deliberately by local farmers who often believe it to be a sensible land use practice in such areas, though often for purely superstitious reasons. At Borth Bog two plants are nearing, or may have reached extinction. These are the bog moss (*Sphagnum imbricatum*) and the brown-beaked sedge (*Rhynchospora fusca*); the latter was once widespread on the reserve but has not been seen there for the past ten years.

At both Glasson Moss and Borth Bog, the fire prevention measures are inadequate. At Glasson, firebreaks around the central area are mown, but this does not prevent the movement of intense fires during dry periods. At both sites there is a case for considering the construction of wide, water-filled ditches around the reserve boundaries. This would be expensive, but may be the only means of saving these vulnerable habitats.

There is also a need for more research into the recovery of peat surfaces from burning and its influence on species composition. Evidently such bogs have suffered some degree of burning in the past and have recovered. It is instructive to inspect the diagrams of exposed peat profiles from Danish raised bogs recently published by Aaby (*Nature*, **263**, 281; 1976) which have charred layers running along certain horizons dating from Neolithic and Bronze Age times. The raised bogs recovered from these traumatic episodes, but what will be the outcome of recurrent, sometimes annual fires?

The Nature Conservancy Council in Britain has the responsibility for acquisition, management and maintenance of Nature Reserves and also for the commissioning of relevant research. Its record on the survey and on the acquisition side is quite good, but its success in the maintenance of acquired Reserves is not outstanding. True, conditions have been unusual this summer, but some of the damaging episodes, such as the burning of raised bogs, are regular, predictable events and the NCC must accept some responsibility for failing to take adequate preventative action in good time. No blame for this neglect can be placed with the regional and field staff who have done everything possible within their present limited economic and physical resources. One cannot help feeling, however, that in the case of mature, self-sustaining ecosystems like the raised bogs the NCC considers its responsibilities to end with acquisition and neglects the assignment of funds to their protection. Let us hope that the sad events of this summer will lead to

the redeployment of money and manpower from the administrative to the field management and research aspects of nature conservation. We need fewer paper clips and more gum boots. □

The blind white fishes of Persia

from a Correspondent

A NEW species of blind loach captured in the Zagros Mountains, Iran, has recently been described by Greenwood (*J. Zool., Lond.*, **180**, 129–137; 1976). In itself this is both interesting and remarkable, but it also represents the successful culmination of a quarter-century of interest in Iranian eyeless fishes by their collector, Mr Anthony Smith.

In 1950 Mr Smith led a party of students from Oxford University on an expedition to Kirman in Iran; one of their objectives was to study the fauna of the qanats, artificial underground aqueducts which convey water from the foothills of the mountains to the plains villages. In particular, they hoped to capture fishes which literary sources suggested were blind and depigmented and lived in the channels. On the expedition's return, Smith published a most entertaining account of their adventures (*Blind White Fish in Persia*, Allen and Unwin, London; 1953). Unfortunately, and despite this title, the expedition failed to catch any blind fish (although apparently fishes were found in the qanats they were not described or identified).

In the interval between planning the expedition and its accomplishment, however, Bruun and Kaiser (*Dan.*

scient. Invest. Iran, **4**, 1–8; 1950) published a description of *Iranocypris typhlops*, found in 1937 in a subterranean water system outlet in the Zagros Mountains in southwestern Iran.

In 1976 Anthony Smith returned to the area, and visited the well-like outlet in which *Iranocypris* had been collected. Here he caught three colourless fishes, two of which he brought back to Britain. One of them proved to be *Iranocypris typhlops*, the other, surprisingly, was a loach new to science which Greenwood has just named most appropriately in honour of its collector, *Noemacheilus smithi*.

The loaches are freshwater fishes of wide distribution in Eurasia (although they are also found in north-east Africa); they belong to the order Cypriniformes which also includes the carp-like fishes. Although numerous species are known, particularly in the Asiatic region, this is apparently the first typical cave-dwelling noemacheiline loach to be recognised. Earlier authors had discussed loaches in Assam and an unspecified part of India which had been found in caves, but in neither case were the specimens completely depigmented, nor were the eyes greatly reduced (although there seems to be uncertainty as to whether the latter population showed eye reduction at all).

Noemacheilus smithi, however, is as typical a cavernicolous fish as any described. Its eyes are completely absent (as far as macroscopic examination could establish); the orbit on dissection appeared to be a shallow depression filled with minute fat globules. In life, its coloration is dead white suffused with pale pink, the viscera are visible through the abdominal wall, and the redness of the gills can be discerned through the gill

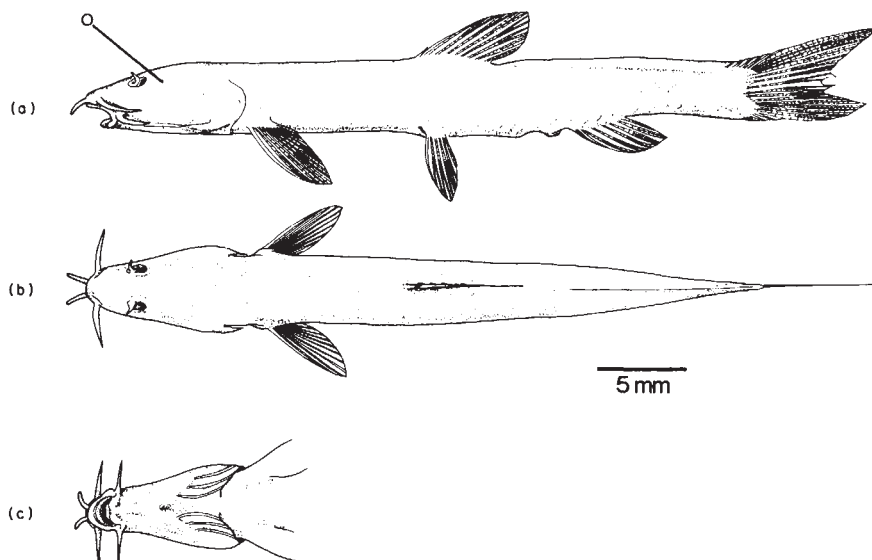


FIG. 1. *Noemacheilus smithi* sp. nov. holotype. (a) Lateral view. (b) Dorsal view. (c) Ventral view of head. O = position of orbit.

cover. In other respects it is similar to other noemacheiline loaches, but the body is more slender than in most species, and there is a long, rather deep adipose ridge along the back behind the dorsal fin. The barbels around the mouth are, if anything, shorter and thicker than in other related loaches.

In a sense this discovery whets the appetite for more exploration in the area. The site in which both blind fishes were discovered is a well-like outlet, fully exposed to light, and in no way a cave. This poses the question, where are these fishes really living? Presumably the well is in contact with a subterranean water system in which both blind fishes live in some numbers. This, as yet, has not been discovered, but it could prove of great interest as this site where two fully-adapted cave fishes co-exist is only paralleled in the Mammoth Cave, Kentucky, a well explored system. Perhaps future exploration will yield further cavernicolous animals in Iran.

Iran's Department of the Environment, so helpful to Smith on his journey, has sent biologists to the area twice to collect further specimens. They found two more of the previously undescribed loach, which are currently being examined at Colorado State University, and plan a further expedition to the cave outlet this winter when water levels rise at the beginning of the rainy season. □

The theory and practice of star formation

from Ant. Whitworth

A Symposium on Star Formation was held by the International Astronomical Union at the University of Geneva on September 6–10.

STARS have formed in the past, and are continuing to form. Of this much we could assure the puzzled Huckleberry Finn who pondered "whether they [the stars] was made, or just happened": but of little more. Ultimately, self-gravity must drive the enormous contractions which convert diffuse gas into new stars. But the critical circumstances of this contraction—as the material destined for stardom overcomes the inhibitions posed by the internal thermal pressure, magnetic stresses, centrifugal acceleration, tidal disruption and turbulence—are extremely uncertain. The standard belief

is that most stars form in clusters, and the primary aims of star formation theory are then to predict, first, the distribution in mass, and in phase-space, of stars formed in a cluster, and second, the evolution in time of the external appearance of a star cluster during formation. To this end, four stages might usefully be distinguished: (1) the creation of a massive proto-cluster cloud; (2) the separation of individual protostars from such a protocluster cloud; (3) the contraction of a protostar to form a star, and (4) the interaction of a newly formed star with surrounding residual matter until the star becomes visible at a distance as a nonaccreting, unobscured, main-sequence star. (This fourth stage is included because it provides almost all the observable evidence of star formation).

At the symposium it was clear that, although the recent flood of observational data throughout the electromagnetic spectrum has tried many established star formation theories and spawned several new theories, much work lies ahead before star formation will be understood. For instance, the role of a spiral density wave and its associated galactic shock in initiating formation is very uncertain: whether it be to force a phase-change on the interstellar gas, thereby converting intercloud gas into clouds; or to assemble massive protocluster clouds by running together pre-existing small clouds; or simply to trigger by compression the gravitational collapse of pre-existing massive clouds. If we introduce the equally uncertain—and presumably coupled—role of magnetic flux, cosmic rays and their associated instabilities, we have already posed a long chain of unanswered questions and are still a long way from forming stars. B. Bok (Steward Observatory, University of Arizona) pointed out that there is extensive continuing star formation in the Magellanic Clouds; nowhere is the evidence more clearly laid out in the sky for the observer; yet there is no clear indication that star formation is very intimately involved with spiral structure, let alone an associated shock.

If such questions are put aside, and the existence of a gravitationally bound protocluster cloud is adopted as a starting point, we confront the problems of separating out individual protostars: by fragmentation or condensation, accretion or accumulation. Are protostars formed by an hierarchical fragmentation sequence which breaks the protocluster cloud up, in a cascade towards increasingly small, dense pieces, as originally envisaged by Hoyle? More probably, contemporary protostars condense directly out of a relatively low background density

($n \lesssim 10^4 \text{ cm}^{-3}$) due to gravothermal instability. Certainly no clear evidence for hierarchical fragmentation has been found in radio line (for example, CO) mapping of molecular clouds, in spite of the use of ample resolution—although any information would surely be a complicated convolution of density, temperature and optical depth structure. The only clear observations of fragmentation during star formation are (1) the high incidence of binary and multiple star systems, which implies that the inception of a typical protostar occurs in circumstances dominated by angular momentum, and (2) young clusters compounded by OB associations.

C. Lada (Center for Astrophysics, Harvard University) and B. Elmegreen (Princeton University Observatory) drew attention to a significant pattern observed in star formation regions. Ideally, this pattern is a regularly spaced alignment, parallel to the galactic plane, of first an elongated, quiescent molecular cloud; then an excited molecular cloud exhibiting signs of internal activity and star formation (infrared sources, masers and compact radio HII regions); next a compact OB association collectively exciting an extended optical HII region; following this an expanded OB association; then an even more expanded OB association. They submit that this sequence may arise as follows. An OB association formed in the front end of the cloud excavates an HII region, and sends an ionisation front back into the cloud. This ionisation front is preceded by a shock front, and after a time there is a shocked layer of neutral gas between these two fronts, which is sufficiently dense and cold for a new OB association to form by gravitational instability. And so the process repeats itself with a period of $\sim 2.5 \times 10^6$ yr, producing a string of OB associations at intervals of ~ 15 pc.

Given the considerable uncertainty which surrounds the inception of protostars, it is very difficult to predict the statistics of newly formed stars. C. Low and D. Lynden-Bell (Institute of Astronomy, Cambridge) have made a detailed evaluation of opacity-limited limit of $\sim 0.007 M_{\odot}$ for the local fragmentation and derive a lower mass temporary gas. J. Silk (University of California, Berkeley) has reproduced the Salpeter initial mass function (IMF) on the basis of opacity-limited fragmentation, and has also made several other quite definite predictions which occur with observational inferences: small stars form first at the centre of a protocluster cloud and heat up the outer layers, thereby increasing the Jeans' mass; consequently massive stars form later near the cloud surface.

Formation of the most massive stars,

however, is commonly assumed to involve an appreciable accretion phase. The upper mass limit is then attributed to an accretion cutoff, when the underlying star develops a sufficient ratio of luminosity to mass, that radiation pressure (acting through grains) halts the inflow. S. Strom (Kitt Peak National Observatory) suggested that accretion may instead be halted by a strong stellar wind. But the essential point is that Silk's IMF for fragments may not be completely relevant if the mass of the final star is much less than that of the fragment, the balance being contained in an unaccretible cocoon, as envisaged above (or perhaps in a disk which serves as a sink for the star's unwanted angular momentum). The final mass may then depend more strongly on the critical accretion parameters (density, opacity and velocity dispersion in the ambient gas).

Moreover, simple arguments show that if, at its inception, the protocluster cloud contains realistic measures of frozen-in magnetic flux and minutely conserved angular momentum, then subsequent contraction and fragmentation will amplify magnetic and centrifugal stresses until they assume dominant roles in the dynamic evolution. I. Mestel (Astronomy Centre, Sussex) pointed out that until—or unless—the charged particle density be-

comes very low, the gas is coupled to the magnetic field, and so clouds must tend to contract along the local field-lines; if the gas remains coupled to an attached, large-scale field for long enough, magnetic stresses may transport angular momentum from a cloud to its background, thereby postponing the development of centrifugal stresses until individual stars approach the main-sequence; by this stage the gas is recoupled to the field, and magnetic braking may again act to slow the rotation of the newborn star.

In conclusion, existing theory can say nothing about the overall efficacy of star formation. So many possible contributing processes can, at best, be evaluated in terms of their energetic feasibility, and seldom in terms of their competitiveness. Observations indicate that star formation in the solar vicinity presently converts ~5% of the interstellar medium into stars at each passage through the spiral pattern. Various attempts have been made to fit the mean rate of conversion of interstellar matter into stars with a law of the form $d\ln(\rho)/dt \propto \rho^n$, where ρ is the mean space density of interstellar matter, and n is to be determined by fitting observations. The results are conflicting and probably no such simple relationship exists where star formation is concerned. □

ozone chemistry is not a theoretical myth but a reality in the stratosphere. It is not clear, however, how much of this observed chlorine comes out of the CFMs 11 and 12 and how much from natural sources like CH_3Cl , CCl_4 and so on; what are the other competing reactions in which Cl atoms get involved to make them inactive for ozone reduction; and lastly, how many of them are removed from the stratosphere before they meet the ozone molecules. These are the uncertainties which led the NAS panel to put a factor of 10 on the uncertainty in their estimate of ~7% for the eventual reduction of ozone by CFMs. This implied that the eventual reduction of ozone could be anywhere between 2 and 20%. Several of these uncertainties were discussed at the conference. It appeared, for example, that the removal of Cl by the production of and eventual precipitation of the ClONO_2 molecule may not, after all, be such an efficient mechanism. Other uncertainties, however, remain. One hardly expected them to be answered only two days after the release of the NAS report. But one thing became very clear; with the amount of scientific activity in this field, this time next year the uncertainties will certainly be reduced by a significant amount.

The other scientific issue discussed at the conference was that of N_2O ; its sources and sinks near the ground and its eventual transport to the stratosphere. The problem, if it exists, could be greater than that of CFMs. It appears that N_2O , a product of biological denitrification in the soil, does not get easily "rained out" in the troposphere, diffuses on to the stratosphere, reacts with atomic oxygen, gets converted to NO and NO_2 , which then acts as a catalytic agent to destroy ozone in the upper atmosphere. Does this really happen or not, is of course an interesting scientific question for the students of ozone chemistry in the stratosphere. But the problem becomes greater when it is pointed out, as was done by Michael McElroy of Harvard last year, that the ever-increasing use of fertilisers may considerably accelerate the release of N_2O to the atmosphere with an eventual impact on ozone much greater than that predicted for supersonic aircraft, or the CFM.

Is this a real problem? We do not yet know. First, one needs much more precise information on the sources, sinks and life time of N_2O in the "natural atmosphere". What are the fluxes over the land and over the oceans? How much is the air-sea exchange? What is the life time of N_2O in the troposphere? What is the fixation time on the surface? Again, as in the case of CFMs, the theory seems

Stratospheric decision making

from S. I. Rasool

An International Conference on the Stratosphere and Related Problems was held at Logan, Utah on September 15–17, 1976.

It is refreshing to come out of a two and a half day international conference with a certain sense of accomplishment. This was partly due to the theme of the conference: we not only discussed the physics and chemistry of the stratosphere, but one day was dedicated to questions related to the dynamics of decision making on the stratosphere. Nowadays when the stratospheric scientist has suddenly become an important individual, to both the media and the policy makers, interaction between the three was productive and satisfying, helped along by well-chosen speakers and panel discussions in which the major issues raised at each session, were if not solved, at least clarified.

In particular the conference took place two days after the much awaited release of the National Academy of

Sciences (NAS) report on the CFM problem. (*Nature*, **263**, 268; **263**, 723; 1976). After this conference and after the report where do we stand on the question of stratospheric impact of man-made products and what should be our attitude towards "regulation" of these products?

As for chlorofluoromethane (CFM) release, much progress has been made since the publication of the now famous paper by Rowland and Molina (*Nature*, **249**, 810; 1974). We now know that most of the CFM 11 and CFM 12 ever released has accumulated in the atmosphere. We also know that a few per cent of them are reaching the stratosphere. There is also evidence that they are being dissociated, producing chlorine atoms which have a potential of catalytically reducing O_3 to O_2 . A major new set of measurements reported by J. Anderson (University of Michigan) confirmed, for the first time, the existence of Cl and ClO in the stratosphere. This was a crucial measurement that actually demonstrated that the chlorine-

to be in hand but the crucial measurements are just beginning to be made. As the Chairman of the Session, H. Schiff, put it: "We don't yet know whether the laughing gas problem is a 'laughing matter' or not".

The most exciting session of the conference was of course the one dealing with the dynamics of decision making and the topic under discussion was whether to "regulate" or "not to regulate" the use of CFMs 11 and 12 although the scientific evidence of the magnitude of their impact on ozone is still largely inconclusive. Policy makers from Washington, including the Chairman of President Ford's Council of Environmental Quality, discussed how decision making on issues of public policy proceeds in the face of scientific doubt.

About two million man-made chemicals now exist, and about 25 thousand are added each year. Since it is impossible to deal with them on a case-by-case basis, some general policies and philosophies are obviously needed. Those chemicals which have direct and immediate effects such as food poisoning or acute radiation exposure are of course easy to handle. For those chemicals whose effects are not immediately obvious and may occur several years or decades later, such as cancer-inducing products, or the CFMs, decisions on control have to be taken from a different perspective. In these cases public concern is not always an accurate indicator of acceptable risk and one has to keep in mind that often the values of the lives of the yet unborn are not assumed to be the same as those of our own. Sometimes it may also be unwise to postpone the decision until the effects can really be measured because a major impact may already have been made.

How then should we proceed? It was abundantly clear in the minds of the policy makers that although there remain valid doubts about the effects of CFM on the ozone layer, from the standpoint of public policy there remains no valid reason to postpone the start of regulatory procedures on the use of CFMs 11 and 12. They strongly suggested an immediate voluntary phase-out of non-essential uses of CFMs. This opinion was in direct conflict with that in the NAS report calling for a wait of up to 2 years depending on when the uncertainty can be reduced. The Academy panel was severely criticised for going beyond its charter and pronouncing judgement on a public issue. "There are no individuals better qualified to make scientific judgements than scientists. But scientists are no more qualified men than anyone else in making social value judgements," said Russell Peter-

son. "Moreover, this judgement carries with it added credibility simply because NAS made it and sets a strong public mood," added David Pittle, Commissioner of the US Consumer Product Safety Commission.

The debate between the scientists and the Washington bureaucrats was certainly educational for both. The scientists were reminded of their responsibility to present complex issues so that they could be understood by the public and that the uncertainty in their predictions is more evident than obscure. They were urged to place their own work (and bias) in perspective so that the media and public get a balanced view of the issues and answers.

For the scientists, so used to peer review and refereed journals, it was perhaps a little difficult to understand how one really assesses the importance of public opinion which is at once so volatile and so dependent on the media coverage of the news. We were assured by the Environmental Protection Agency that there were cost benefit analyses before any economic judgement was made and we were told that several months of public hearings take place before a judgement on the public opinion is made.

This conference was a satisfying affair. I think we all came home a little more reassured about each other and about each other's role in the eventual solution of the problem. □

Earth's core in Oregon?

from Peter J. Smith

LAST year Bird and Weathers (*Earth planet. Sci. Lett.*, **28**, 51; 1975) made the startling suggestion that josephinite, a rare rock consisting largely of nickel-iron alloy, may have originated in the Earth's core, or at least in the vicinity of the core-mantle interface. If this view were to be substantiated it would be of more than passing interest, for proven examples of core material in the crust are not exactly common. It would also carry with it certain implications about the Earth's ability to transport material over great vertical distances. Indeed, Bird and Weathers proposed specifically that josephinite may have been raised from the core-mantle interface in a mantle plume, carried into the lithospheric mantle by diatremes and emplaced into the upper crust by ophiolite obduction. But none of this exoticism was destined to go unchallenged; and Dick and Gillete (*Earth planet. Sci. Lett.*, **31**, 308; 1976) have now taken Bird and Weathers to task on a number of points which taken

together suggest a much less spectacular origin for josephinite.

The basis of the case put by Bird and Weathers was that the josephinite found as pebbles at Josephine Creek, Oregon, is distinct from the more common, albeit superficially similar, mineral awaruite. The chief differences claimed are that josephinite is much coarser-grained than awaruite and that, unlike awaruite, it contains two metal phases (nickel-iron alloy and α -Fe) and andradite garnet. Moreover, some josephinite samples also contain elemental silicon and a natural variety of the artificial substance CaO. 2^{+}FeO^{+} , neither of which has previously been found in terrestrial rocks. Such differences were held to support the view that josephinite and awaruite have a different origin.

But even some of the facts are in dispute. Apropos of the reported discovery by Bird and Weathers of two metal phases, for example, Dick and Gillete claim that the metal in josephinite is all nickel-iron alloy, albeit with a "large bimodal compositional variation" between samples. There are indeed two metal phases, but one is awaruite and the other is taenite (rather than α -Fe), both of which are nickel-iron alloys. Dick and Gillete do admit that the association of the two metal phases with andradite in the samples analysed by Bird and Weathers is uncommon, although they point out that many of the pebbles contain only awaruite. They also quote a forthcoming report from Botto and Morrison which claims that there is in fact a complete gradation between awaruite and awaruite-taenite-andradite.

Next, Dick and Gillete throw doubt on the presence of elemental silicon in some samples which can be interpreted in other ways. In any case, they question the view of Bird and Weathers that the presence of silicon would preclude origin in the lithosphere, citing the production of impurity silicon in the smelting of iron as evidence of the element's ability to form stably at low pressures. The suggestion that andradite garnet/nickel-iron is not a stable assemblage (implying that the two components could not have formed together) is also disputed, largely on the grounds that Gustavson's (*J. Petrol.*, **15**, 455, 1974) experiments on the stability of andradite have been interpreted by some of his successors too uncritically and incompletely. And finally, Dick and Gillete suggest that the presence of diamonds in Josephine Creek, even if correctly reported (it is in fact in dispute), does not necessarily support the case that Bird and Weathers were making, if only because diamonds are representative of upper, rather than lower, mantle pressures.

Also relevant to that case are the dif-

ferences, or supposed differences, between the nature of the pebbles (which are placer deposits) and that of similar *in situ* material within the mass of the nearby Josephine Peridotite. The Josephine Peridotite, partly unaltered harzburgite and partly serpentinite, is apparently a segment of ophiolite emplaced by obduction. All parties agree that it contains awaruite in parts and that awaruite is formed by the serpentinisation of ultramafic rocks. They likewise agree that the Josephine Peridotite is the source of the pebbles.

But the consensus then breaks down. Dick and Gillette claim that both the *in situ* material and the pebbles are awaruite and (bearing in mind the nearness of the pebbles to a zone of intense shearing, serpentinisation and igneous intrusion within the Josephine Peridotite) that both are the products of low-temperature hydrothermal metamorphism and serpentinisation of the peridotite. Such differences as there are between the two (for example, it is acknowledged that the pebbles have unusually large grain size) represent varying conditions imposed by igneous

intrusion during serpentinisation.

Bird and Weathers, on the other hand, claim that the differences between the pebbles and *in situ* material are serious enough to imply a different origin (and to require separate names) for the two. Having acknowledged that the source of the pebbles is the Josephine Peridotite, having agreed that the *in situ* awaruite is the result of serpentinisation and having proposed a core origin for the pebbles, this would appear to put Bird and Weathers in the position of saying that the Josephine Peridotite once contained (and perhaps still contains) two distinct but closely related nickel-iron alloys whose zones of origin were separated by more than 2,000 km vertical distance.

Obviously it is possible. For example, a mantle plume could have been feeding the accreting plate margin from which the Josephine Peridotite is derived. But is it credible? If so, the exciting possibility of core material at the Earth's surface remains. If not, the balance of the argument must be in favour of Dick and Gillette who in any case have Occam's Razor on their side.

details of a number of sophisticated applications in nuclear physics. Following on from this, M. Loponen (Otaniemi) introduced a lively debate on the problems and pitfalls of secondary thermometry.

One of the most fascinating, and perhaps also the most important, topics of the conference was reserved for the final day when K. W. Taconis (University of Leiden) and F. A. Staas (Philips, Eindhoven) talked about the new cycle in which instead of extracting ^3He from the dilute phase in the mixing chamber using a still, ^4He is pumped in through a superleak. This technique offers the great advantage that the dilute and concentrated phases counterflow in the same tube, rather than in separated tubes, so that no heat exchangers are needed. The simple Leiden machine, which reached 8 mK very soon after having been commissioned, was discussed in detail. Hybrid machines in which a "Leiden" mixing chamber is piggy-backed on a conventional refrigerator, and both ^3He and ^4He are separately circulated, as developed at Philips and Grenoble, were also considered. The meeting ended with an all-too-short session presided over by E. J. Varoquaux (Orsay) on the actual practical problems associated with operating refrigerators.

Notwithstanding the astonishing progress of the past 10 years, the dilution refrigerator is clearly still capable of significant improvement. If, as one suspects, commercial machines capable of reaching 5-6 mK soon appear on the market, it will be very largely a result of the beautiful research and development work currently in progress on this side of the Atlantic. □

Millikelvin technology

from P. V. E. McClintock

A Europhysics Study Conference on Dilution Refrigeration and its Applications was held at Lancaster University, UK, on September 25-27 1976. The meeting was organised by D. Thoulouze (Grenoble) and G. R. Pickett (Lancaster).

It is now more than a decade since the first dilution refrigerators were built. During this period the machines have become the standard means of reaching temperatures between 10 and 300 mK in a continuously operating mode and, moreover, the technique has spread far beyond the area of purely low temperature physics and is being widely used in a number of other fields.

Dilution refrigerators operate on a mixture of liquid ^3He and ^4He making use of the property that, at low temperatures, the liquid separates into two phases, usually known as the concentrated phase and the dilute phase. The concentrated phase consisting of almost pure ^3He floats on top of the dilute phase, which is composed of a gas-like assembly of ^3He atoms moving freely in a "background" of superfluid ^4He . Thus the movement of ^3He atoms from the concentrated to the dilute phase is very much like the evaporation of an ordinary liquid, and is

accompanied by a similar cooling effect.

The conference started with an introductory session led by H. E. Hall on what has become accepted as the conventional cycle: the "General Motors, rear-wheel-drive refrigerator", as he called it. In this cycle, ^3He is drawn out of the dilute phase by a heated still and the cooling effect arises from a balancing flow from the concentrated phase. The process is sufficiently well understood that machines capable of reaching 10-12 mK are available on the commercial market. The two sessions which followed led by G. Frossati (Grenoble) and A. Th. A. M. de Waele (Eindhoven), discussing the optimisation of this technique, provided what in many ways formed the high spot of the conference. It was particularly impressive to hear that the machines developed in Grenoble are able to reach 4 mK (not very far above the liquid ^3He superfluid transition) as a matter of routine and, furthermore, that their performance is in reasonable agreement with calculation.

On the second day some of the recent practical applications of dilution refrigerations were described in sessions led by R. E. Packard (University of California, Berkeley) and T. O. Niinikoski (CERN), and included



A hundred years ago

RUSSIAN newspapers announce the death of M. Chekanoffsky, who, exiled in Siberia, has spent more than ten years in the geological exploration of the country, and recently returned from his travels on the Olenek and the shores of the Polar Sea, to St. Petersburg, where he was engaged at the Academy in the description of his immense collections. He was found on October 10 dead in his room, and it is supposed that he poisoned himself. From *Nature*, 15, November 9, 50; 1876.

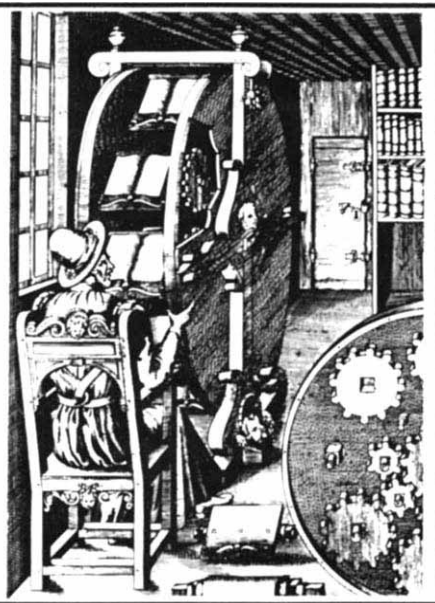
WHATEVER happens now, we cannot blame the Marxists for demystifying science. It's true that they were first in the field in England, shocking the bourgeois at the British Association at Durham in 1970 with their slogan: "Science is not neutral". But the point was soon reiterated to greater effect, not merely by Lord Rothschild's gauntlet, but even more by Sir Frederick Dainton's intended smooth covering. It was he who mentioned a "dangerous and corrupting 'ivory towerism'", to be countered by a continual awareness of "national needs and objectives" by those planning basic and strategic science. And since then we have all learned that the language of science is priorities, with a grammar of shrinking budgets.

Of course, the points are made with quite different motives from the two sides: our established leaders want more 'civic responsiveness' from science, while the counter-slogan is 'social responsibility'. And the analyses of the situation of science diverge quite radically. Those who would manage science act on behalf of a social order assumed to be basically good, and capable of further improvement by the proper application of science. The Left critics exhibit science as a microcosm of an unjust society, and expose its hypocritical pretensions to superiority.

Indeed, the general impression that may be conveyed by the new double-decker edited by the Roses, *Ideology off/in the Natural Sciences*, is that of a many-sided assault on cherished illusions. For them and their collaborators science is neither innocent, ennobling, or even nice. The effect on a thoughtful scientist could well be similar to that on a benevolent, complacent male who picks up the *Spare Rib* that his women-folk avidly read, and discovers what he looks like from below. Although he may consider himself eminently fair in playing the game of social life, he is there told that the rules are rigged, for him and against the others. And if he is smitten with concern and guilt, he also learns that there is no easy cure for maleness as a social disease.

Similarly, those who enjoy the benefits of participation in the upper reaches of the institution of science are typically white and middle-class as well as male; thus they belong to a group whose security, until now enormous, is now decreasing markedly. Moreover (and this is an important thrust of the book), the character of the class of scientists is closely bound up with that of the science that they produce or apply. In its objects, methods and functions, our science is a creature of its matrix in a particular sort of class society. Stripping away the ever thinner veneer of 'pure science', these critics find that science is supported for its

Book review supplement



contribution to two ends: production, industrial and military, through physical inventions and discoveries; and social control, through its results, methods and above all ideology.

In this last point the authors initiate a major advance in Marxist thought.

Assault on cherished illusions

J. R. Ravetz

Ideology off/in the Natural Sciences. (Two volumes.) Edited by Hilary Rose and Steven Rose. (Macmillan: London, November 1976.) *The Political Economy of Science*. Pp. 320. *The Radicalisation of Science*. Pp. 260. Each volume: hardcover £10; paperback £3.95.

Until now, the Marxist tradition (along with positivism and rationalism) has assumed natural science (as best exemplified in physics) to be above the contradictions of society and independent of false ideology; from that firm basis, intending revolutionaries could proceed with certainty to transform the social world. The authors here are determined to exhibit the socially conditioned ideology 'off/in' natural science, without collapsing into relativism or irrationalism. This is a task of some delicacy, here only begun.

But a beginning there must be, and the authors have provided it, mainly (and properly) by accounts of areas they know from their own experience. These books have advantages and drawbacks of a largely amateur collection; there is repetition between essays

(as well as divergence of approach and doctrine), some looseness of organisation within them, and occasional bursts of rhetoric not intended for most of the readers of this present review. But generally, they are refreshingly free of jargon and bombast. And there are many insights to be gleaned in them; I shall try to convey some of these.

To accomplish this thoroughgoing critique, there must be a very broad definition of 'science', really encompassing all of theoretically trained problem-solving expertise. The most visible part, academic 'pure' natural science, is in this analysis reduced to a small, self-deluded sector. In one of their more breathtaking asides, the Roses remark: "By the end of the war, the autonomy of science had become a myth helping to ensure that those students with first-class degrees remained in the universities and those with seconds went into industry". (*Radicalisation*, p7.) Cruel, to be sure; but is it a complete caricature? And since there is no consensus on the boundaries of 'science', one must judge any proposed definition by the effectiveness of its use.

The Roses and their co-workers are not just demystifying and muck-raking. They are proclaimed Socialists, taking their inspiration from Marxism (although in what they call its "problematic inheritance"). Their essays are then both a criticism of capitalist natural science, and a demonstration of Marxist social analysis, as enriched by themselves. Their Marxism is the opposite of dogmatic: indeed it is easier to say precisely what it is *not* in terms

* Illustration of the "Book Wheel" taken from *The Various and Ingenious Machines of Agostino Ramelli*. Translated with an introduction by Martha Teach Gnudi and Eugene S. Ferguson. Pp. 604. (Scolar: London; John Hopkins University: New York, July 1976.) £50.

of the worries it relegates (environmental problems, and community and personal development), than what it is, by the articles of belief it maintains. Even the placing of the authors with respect to movements and powers in the contemporary world is rather vague; the book is dedicated to the "Heroic Peoples of Indochina", and there are occasional exhortations to workers; but otherwise it is all somewhat abstract. Even their citations to professedly Marxist literature on their subjects are very sparse. This independent policy has its benefits, of course; the authors are then entirely free to criticise capitalist culture on the basis of its own best values of liberty, decency and honesty—rather like Marx himself. Also, it enables them to move implicitly in some very interesting directions, of which more later.

What, then, can scientists see of themselves through this radicals' mirror? The Roses' leading essay "The Incorporation of Science" gives a synaptic view of the field (as their introduction gives admirable summaries of all the papers). The new conception of science, first described simply as 'big' but now as 'incorporated' or 'industrialised' subverts all the ideals of nineteenth century 'pure' science, and also many of those of the 'improving' tradition from Bacon to Bernal. But most academic studies of science (history, philosophy, sociology) are as yet only dimly aware of this transformation "from scientific community to scientific factory". Perhaps this is because they relate, not to the mass of *scientific workers* who are found to be indifferent to the hallowed 'norms' of science, but to the visible elite. This group can be identified; some 0.01% of the American scientific work-force of 2 million are the real decision-makers. And it is in their interest to maintain the myths of autonomy and 'public knowledge' as the ruling ideals of science. With this demystification accomplished, the book is launched on *The Political Economy of Science*.

An analysis of modern science-based industry that is broadly in line with classical Marxism is provided by Mike Cooley. He describes the proletarianisation of white-collar and technical labour, along with the continued dehumanisation of production-line work through Taylorism. His own experience in AUEW-TASS provides thought-provoking examples: computer-aided design, distorting the balance between the quantitative and the qualitative and leading to a stifling machine-enforced bureaucracy in the design process; attempts to restrict factory recruitment to men under 30 (they 'burn them up' in 10 years); the series of three strikes necessary to get heating and ventila-

tion for workers comparable to that provided for computers. This ultra-high technology industry aggravates many of the contradictions of capitalism analysed by Marx and also has created a few of its own: the rapid obsolescence of knowledge, and the extreme vulnerability of such systems to 'guerilla'-type action—be they works to rule at home, or Sino-Vietnamese warfare abroad.

The attempt to explicate science (as distinct from science-based industry) in orthodox Marxist terms is more demanding; a group of Italian physicists make a brave try, but it is not very illuminating for those not sharing the paradigm. The second half of their essay, based more on their personal experience, is more rewarding. By contrast, the less theoretically-orientated analysis of André Gorz provides a comprehensive radical sociology of scientific knowledge. He sees the system of scientific research from the outside, and accordingly can make criticisms that would be difficult for an established research scientist, however rebellious, to express.

For Gorz, all the features of science that render it the preserve of an upper-class minority, are solely to be explained by their social functions. Science itself is effectively defined by formal teaching and diplomas; traditional or self-taught craft skills have no standing against certified expertise. And only those "socially qualified to exercise authority" will succeed in the training system; thereby ensuring that this knowledge is not utilised by the wrong power. But in any event, most of the knowledge of science is alienated and irrelevant to life, in a unique degree. The people are totally excluded, and the experts are fragmented and helpless (and in the case of academic researchers, useless as well). It is then easy for science to be directed to answer some questions and ignore others (Gorz here mentions high technology medicine squeezing out old traditions and communal self-help). Merely changing ownership relations seems to make little difference, so long as knowledge (and the associated power) is hierarchical. Indeed, in passages that recall Rousseau and the Jacobins rather more than Marxism, Gorz rejects *all* science that cannot be shared among 'the people' as their own.

The discussion of 'ideological' conditioning of science starts with fields studied over past years by the Roses: IQ and 'biologism'. Perhaps because of one's prior familiarity, the feeling of "yes, but" increasingly obtrudes on one's reading of the well-argued essays. For example, we are told explicitly that theories linking race and IQ "provide an apparent scientific rationale for the existing social order", almost as if our rulers wanted or called for it. But the directors of our educational system

(surely an area of ideological formation and social control) have quite recently jettisoned the IQ as a means of selection, and this is nowhere mentioned. It is generally much more difficult for a radical critic to explain the virtues of a system than its vices; but to ignore the problem is not productive, nor even consistent with the authors' own standards. I must also remark that in the essay on 'biologism', with its catalogue of horrors of behaviour control from the USA, there is no discussion of the current Soviet conception of schizophrenia as a simple mental disease with obvious behavioural symptoms, including, of course, political deviance.

The really 'soft' natural sciences, such as reproduction technology and 'political ecology', are easy targets for analysis, but the studies here are not trivial demystifications. The real contradictions will not be removed by a reduction to 'class' terms; indeed, traditionalist Marxists fare as badly for their ecological blindness, as do the technical fixers of contraception, for their male chauvinism.

This Marxist approach, however, can really prove its power only in a case where the ideology is *not* apparent, where 'objectivity' seems to reign. Such is the case in physics; and J. M. Levy-Leblond delivers a broadside on that

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noble discipline. Of course, one finds factory research there (as at CERN), the sometimes vicious competition for degraded honours as the Nobel prize and the strange cult whereby scientific eminence is a token of practical wisdom. More than this, the author gets into the subject itself, noting its glorification of brainwork over handwork, the pure over the practical, the 'hard' and abstract over the 'soft' and particular. When this science is taken as the paradigm (as for most of this century), its reinforcing of social and sexual elitism, in the educational system and generally, is very strong. Wisely, the author shies away from discussing the relations of this ideology to the 'truth content' of physics; but for a demonstration of how deeply ideology *can* penetrate, this essay is a very good start.

When we come to *The Radicalisation of Science* the contradictions in the authors' program appear clearly. The Roses have a delightfully self-critical history of British Society for Social Responsibility in Science and its associated movements; but they would not pretend that this movement ever achieved any numerical strength, nor that it is likely to do so in the foreseeable future. In the most erudite essay in the collection, the American radical geneticists, Lewontin and Levins, grasp

the nettle of the Lysenko episode, which did so much damage to the Marxist movement among scientists after the war. They display the many factors that made Lysenko plausible at first: not merely the crisis in food supply, the institutionalised paranoia about 'wreckers', and the resentment against elite foreign-orientated Mendelian geneticists; but also the harshness and irregularity of the Russian climate, that made 'averages' less meaningful than in milder environments and so required different methods and criteria of adequacy in statistical work. For these authors the essence of the matter was that Lysenkoism was an abortive Cultural Revolution: it went wrong because of the particular causes (and also effects) of Stalinism. This might be interpreted as a cautionary tale for hyper-Left reformers of science; and comparison with earlier populist attempts, as the 'Jacobin science' in the French Revolution and the Paracelsians of the Puritan Revolution, would repay study.

How, then, could the true radicalisation of science be achieved? It is a pity that we are not given a successful case study as from Indo-China or Cuba, where a mainly political approach will have been tried. For otherwise, we are left mainly with the women and the inimitable Joseph Needham. (The moving essay by the American Black scientist tells us more about his problem than about an effective solution.) And quite suddenly, Needham does not seem so eccentric as previously in the company of professing Marxists. Of course, he is still developing philosophically, as young in spirit as ever. Here he takes on a new challenger, the counter-cultural Roszak; and like the great Middle Kingdom itself, absorbs and assimilates him. He does so by moving his own Taoism a bit closer to its mystical, revolutionary tradition.

Given Needham's insights, it is really quite simple: 'our' science (be it capitalist or European), is all *yang* and no *yin*; like our civilisation in general, its maleness is dangerously predominant over its femininity. The women's critique of science fits in perfectly here; one (Couture-Cherki) shows the extent

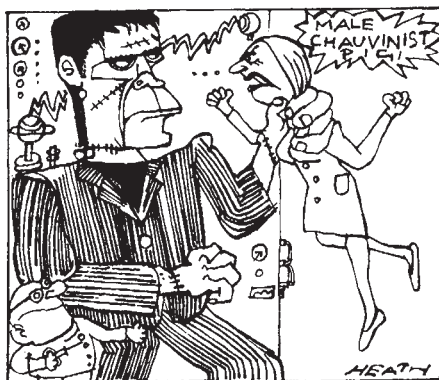
of discrimination and explicit piggery in French physical science; the other (Stéhélin) hints tantalisingly at a feminine cosmology that would enable women to escape the trap of becoming pseudo-males in order to succeed at male-shaped natural sciences. And if this seems fanciful, we can refer back to the analysis of physics by Levy-Leblond.

Of course, no-one has a recipe for *this* sort of radicalisation of science, at least no-one in the Marxist tradition. But is it really a more forlorn hope than transforming science and society by means of an industrial proletariat? After all, Marx *did* believe that ownership relations were the main fetters on the essentially beneficent forces of production; we now cope both with reactionary 'non-ownership' societies and with socially and environmentally malignant commodity production.

This just about brings us back to the worried male (perhaps a scientist!) who has discovered *Spare Rib*. It also may convey a lesson about social change, which is indicated in several essays here. Regardless of how clear or apparently 'objective' may be one's vision of the social world, one cannot simply ignore one's individuality and, from outside, engineer a solution to its problems. For each of us is part of the problem; and in resolving it we must discover (and perhaps demystify) ourselves. Perhaps the next step forward in the study of ideology 'off/in' science will come from the full working out of this principle in connection with the accepted public knowledge of the natural world. In the meantime, these essays make a very good start; and the editors and authors may well be among the first to share in the next move of the game.

But when all this is said, where does such a neo-Marxist critique leave science? In getting from today to tomorrow, science has enough to do nowadays just surviving. It always happens that the more radical the analysis, the more irrelevant it is to ordinary experience, and the more useless it is as a guide to everyday practice. But change goes on everywhere frequently accelerating rapidly. If we are to understand the emerging radical insights about the social activity of sciences, in their content and contradictions, we must pay serious and sympathetic attention to those who are first trying to articulate them. For this instruction, even those who disagree deeply with the Roses and their colleagues will be indebted to them. □

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Methodologies and myths

R. V. Jones

Method and Appraisal in the Physical Sciences: The Critical Background to Modern Science, 1800–1905. Edited by Colin Howson. Pp. vii+344. (Cambridge University: Cambridge and London, September 1976.) £10.50.

"THIS volume constitutes the first collected edition of work so far done in illustrating an important new development in the philosophy of science, 'the methodology of research programmes'". So claims the editor of this collection of seven monographs which exemplify and examine the ideas of the late Imre Lakatos (who is the posthumous author of the opening paper) regarding the philosophy of science. Five of the papers deal with selected case histories, three in physics (kinetic theory/thermodynamics, wave theory of light, and relativity) and two in chemistry (oxygen/phlogiston, and Avogadro's hypothesis).

The self-styled 'importance' of the new development may make the reader bridle: and he may well ask what a research "program" is. It suggests a deliberate plan by an individual or a group, and it ought therefore to exclude those developments in science where there has been no conscious planning; any attempts by subsequent

philosophers to fit such events into a plan will be falsely based. With these prejudicial observations I have attempted, as an experimenter interested in the history of science, to read the book.

As for the philosophy, there is so much unfamiliar jargon that I can only grasp at clues to determine whether or not it is worth mastering. Lakatos discerns four "logics of discovery", of which one is "methodological falsificationism", and his exposition is strewn with terms of comparable ponderosity. So, rebuffed and bewildered, I have retreated to sample two of the case histories to see how his followers have applied his methods in instances where the language and the events are more familiar to me.

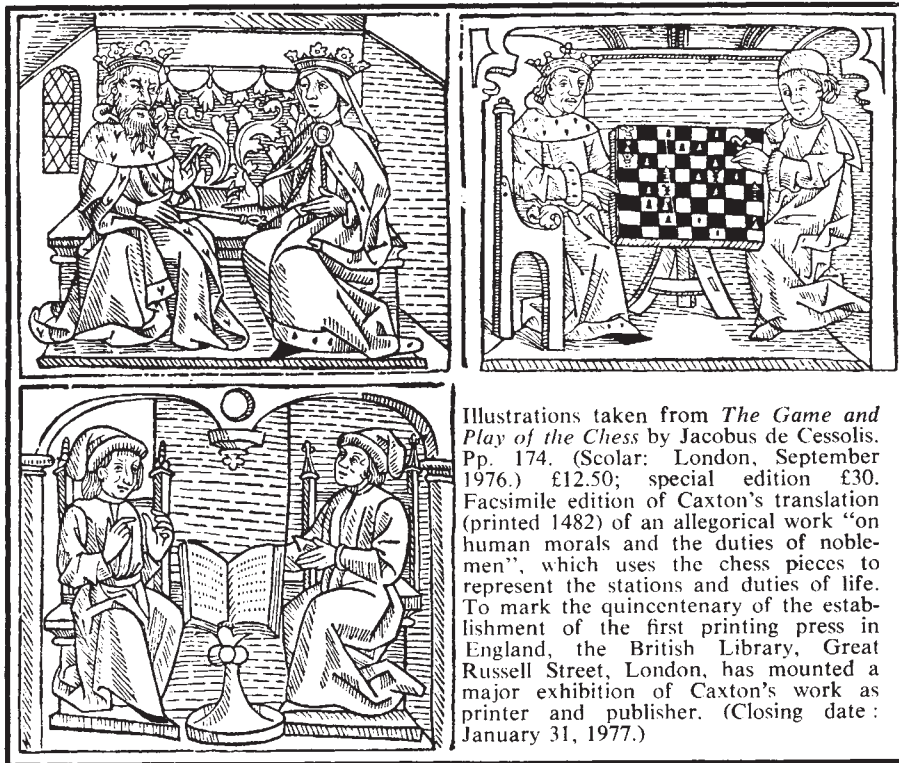
In the course of asking "Why did Einstein's programme supersede Lorentz's?", Elie Zahar claims that his particular ideas of how physics may be furthered by translation into mathematics are illustrated by an account given by Peierls of the way in which Maxwell arrived at his famous equations. Although Zahar credits Peierls with this account (1963) it had appeared in various textbooks of physics long before, and—as Peierls himself took pains to state—there is no evidence in Maxwell's writings that the latter thought, consciously or subconsciously, in the way described. Briefly, the invention of the displacement current is credited to a desire by Maxwell to balance one of his equations of the electromagnetic field. But in his original (1864) paper Maxwell himself gave his

own, quite different, line of thought. He said that he assumed that the aether was a medium of "small but real density" capable of being polarised in a similar manner to a material dielectric; and that while polarisation was taking place there was a displacement of charge which "does not amount to a current because when it has attained a certain value it remains constant, but it is the beginning of a current". Peierls discounts this, by saying frankly that "Maxwell arrived at the extra term by using a picture that we do not accept today". But the question is not what Maxwell might have done if he had present-day knowledge, but by what route he in fact arrived at the outstandingly imaginative concept of a displacement current *in vacuo*. Maxwell himself recorded his approach clearly; and this was by physical analogy rather than by mathematical analysis. So, although most of us are aware that mathematics may suggest creative steps in physics, Maxwell's displacement current is not an example, and its history would have to be bent to fit Zahar's philosophy. Perhaps he is not to be blamed unduly for relying on so eminent an authority as Peierls, but he fails to quote the reservation, made by the latter himself, which completely destroys the argument.

I have to emphasise that I have only sampled Zahar's monograph and it would require much effort to treat it in detail; it has 141 footnotes and 82 references. Peter Clark's study of "Atomism Versus Thermodynamics" runs to 244 footnotes and 196 references, and John Worrall's "Thomas Young and Newtonian Optics", 252 footnotes and 83 references. Such assiduity cannot be reviewed at length, but one further sample will illustrate why to me the book is dangerous. It concerns the famous two-slit interference experiment of Thomas Young.

Worrall says: "There are, I claim, sufficiently many suspicious aspects of Young's account of the two-slit case to support the belief that he never performed the experiment"; and "However, as I have said, there is evidence that Young never performed the experiment"; and once again "Moreover, the dearth of details in Young's account makes it seem unlikely that Young ever did successfully perform it, and certain that he did not give sufficient information about the experiment to ensure its repeatability by others".

What are the facts? Worrall makes play of the point that Young did not give a full account of the experiment, which he described only briefly in his course of lectures at the Royal Institution between 1802 and 1806. Worrall says that when Young really did an experiment he usually gave enough



Illustrations taken from *The Game and Play of the Chess* by Jacobus de Cessolis. Pp. 174. (Scolar: London, September 1976.) £12.50; special edition £30. Facsimile edition of Caxton's translation (printed 1482) of an allegorical work "on human morals and the duties of noblemen", which uses the chess pieces to represent the stations and duties of life. To mark the quinqucentenary of the establishment of the first printing press in England, the British Library, Great Russell Street, London, has mounted a major exhibition of Caxton's work as printer and publisher. (Closing date: January 31, 1977.)

and it is at times challenging. But it supremely illustrates the dangers that now beset the history and philosophy of science. Dionysius may have been right in regarding history as philosophy teaching through examples, and the analysis of case histories is therefore worthwhile on this account. But historians and philosophers of science are becoming increasingly involved in talking to one another in a self-propagating language and literature of

their own, and all too many have no working contact with experimental science, where there is no place for assiduous nonsense. Socrates said that philosophy was not for those without experience: and this is no less important for natural philosophy than it is for moral. □

R. V. Jones is Professor of Natural Philosophy at the University of Aberdeen, UK.

Plasticity and resilience

Freda Newcombe

Early Experience: Myth and Evidence. By A. M. Clarke and A. D. B. Clarke. Pp. x+314. (Open Books: London, 1976.) £3.95. *The Growing Brain: Childhood's Crucial Years.* By John Brierley. Pp. 138. (NFER Publishing: Windsor, 1976.) £3.55.

THESE two books reflect different facets of human growth: early plasticity and enduring resilience. The points of view are reasonable and complementary; the danger is that extracts out of context may be used to bolster overgeneralisations and dogmatic prejudice; and the encouraging aspect is that their authors are searching for the facts—physiological and behavioural—that are necessary for any advance in understanding the complexities of development.

Claims already published about *Early Experience*, ranging from the harm it may do to its "revolutionary implications for the design of education and social policy", obscure its clearly defined brief. The Clarkes have assembled evidence to support a rational hypothesis: that early experiences do not irrevocably determine future development and that "the possibilities for alteration in response to a changing environment remain open for longer than has been commonly accepted" (p18), with the reservation that "this responsiveness decreases in time" (p272). They comment on the inadequacy of received wisdom on child-rearing; they point to the discontinuities in normal development, and the rôle of learning (and—equally important—the possibility of unlearning inappropriate responses); and they note a "remarkable resiliency", a "self-righting tendency" in children, inferred from their physical, intellectual and emotional survival after early years spent in

appalling conditions of isolation and cruelty (see chapters by Davis and Koluchova). Evidence is also drawn from anthropological and animal investigations to show that "the young animal retains an enormous capacity for change in early patterns of behaviour and cognitive competence, especially if the initial environment is seriously altered" (Kagan, p121).

Group studies in the book suggest that the outcome for children initially reared in institutions is related to the adequacy of later placements in other institutions (Skeels) or adoptive families (Tizard and Rees). Children from poor backgrounds may make good progress after relatively late adoption (Kadushin) or institutional care (Lewis; Clarke and Clarke; Reiman). An interesting chapter (Bronfenbrenner) on the so-called "head-start" programs makes the point that the substantial gains shown in the first year tend to fade unless reinforced by follow-up programs and parent participation. The evidence from head-start programs is, however, two-edged. Of more direct impact is the report (Rutter) on parent-child separation, and the significance of genetic and environmental variables. It suggests that the *cause* of separation rather than separation itself may be associated with maladjustment and delinquent behaviour.

The Clarkes have not glossed over the problems of sample, selection and measurement which make quantitative generalisations about child-rearing practices untenable. They are well aware of the constraints on development imposed by constitutional factors; and they do not push their argument to the limit: "there is absolutely no implication that infancy and early childhood are unimportant, only that their long term rôle is by itself very limited" (p272). Negative data, as far as they are aware, do not invalidate a system (p272). By the same token, it is possible that observers have failed to detect subtle changes of language, intelligence, social skills, and personality in children who are at risk in the early years of life. Psychometric tests and questionnaires have their uses and their limits; "the marked

inability to concentrate of many institutional children by the age of five" (p149) is one of the minority notes in this volume that would be interesting to pursue.

It is a pity that some of the authors in the Clarkes' book were recently featured as "the anti-Bowlby team", and that their book has been described as likely to do more harm than good. In turn, the reported suggestion of one of their authors that Bowlby's influence has been "mainly bad" on middle-class mothers is equally inappropriate. It is easy to assign the rôle of spectre in the nursery (*pace* Spock), but that this has perhaps too often been done after somewhat biased interpretations of the maligned experts' texts. And both Bowlby and the present authors have made a positive contribution to the understanding of behaviour. Only those who cling obstinately to a single explanation—the either/or fallacy that clouds so much political and sociological thinking—will adopt a militant stance. The Clarkes' book will be of great interest to those psychologists, clinicians and caseworkers who are concerned with the theory and practise of child-rearing.

DR BRIERLEY'S book, addressed to parents and teachers, summarises work in neurophysiology and developmental and physiological psychology that has a possible bearing on human growth and educational needs. It reflects wide reading in rapidly expanding disciplines and takes on the challenging task of distilling this personal selection into a coherent and feasible view of human development: a brave assignment worthy of Brown-ing's view of man's reach.

Few will quarrel with its main assumption: the child has unique genetic equipment, it is very dependent on the environmental stimulation it receives during the early years of life, and yet it is endowed with a remarkable degree of flexibility and tolerance to stress. The practical implications of this viewpoint are discussed with a plea for an environment rich and varied enough to sustain the diverse needs and talents of human creatures. In contrast, there is little stress on the need for structured training in the early years. In this context, a report in the Clarkes' book that "cognitively structured curricula produced greater gains than play-oriented nursery programs" (Bronfenbrenner, p247) is interesting (a seldom cited and somewhat unfashionable point of view, that might be worth reviving if the percentage of children in the UK with problems in reading, writing and arithmetic is to be reduced).

Given the vast scope of the book,

it would be unreasonable to cavil at details. Inevitably, the selection of experiments will not be approved by all physiological psychologists. Similarly, neuropsychologists may find the functional maps of the brain oversimplified and note that the concept of hemispheric asymmetry of function and the special rôle of the right cerebral hemisphere for *certain* aspects of visual and spatial perception were established long before the study of a small group of patients with callosal sections (p53). Linguists may well be startled by the view that "the remarkable power to speak foreign languages . . . can be explained on the basis of conditioned reflexes" (p 96); and those concerned with brain mechanisms may consider that too much stress has been placed on brain weight at the expense of circuitry and the complexities of modular organisation. The account of physiological

mechanisms and biochemical changes that may underlie the processing and storage of information in the brain is necessarily speculative; and the section on memory and learning is limited to the notion of a "hippocampal taperecorder" (p90), omitting much recent clinical and experimental work in this field.

As a general introduction to the topic, however, the book is a useful, well written and stimulating guide, remarkably free from jargon and pitched at the right level for the readers for whom it was designed. Few would do better or even have the courage to attempt it. □

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Representation of meaning

John Morton

Language and Perception. By George A. Miller and Philip N. Johnson-Laird. Pp. viii + 760. (Cambridge University: Cambridge, London and Melbourne, August 1976.) £12.50.

THIS book is about the representation of meaning. Tired of waiting for linguists to produce a usable semantic theory the authors have started to build a psychological one. The starting point is the perceptual world and its representation in memory. A conceptual theory of memory links this with language. On the one hand we have words and on the other we have ways of testing the perceptual world to find what concepts are appropriate to describe it. In this way we can learn and put what we see or remember into words, or take appropriate action when someone addresses us. In the latter case the sentence is translated into a program which can then be executed appropriately.

Miller and Johnson-Laird illustrate their position with the sentence "Did Lucy bring the dessert?" This is translated into a program which they represent as: find (M(EP), $F(x, y)$). This program will be interpreted as an instruction to search for a particular event in a particular (episodic) memory store. F is a pointer to a particular operator which characterised the meaning of the verb 'BRING'. x and y point to the concepts corresponding to 'LUCY' and 'DESSERT'

respectively. This translation procedure is automatic for the listener who knows English well. He then has the option of answering. This is organised by an executor, which must find the episode and apply the various tests which define BRING: (i) find (domain, COME (agent)) and call it e ; (ii) test (during e , HOLD (agent, object)).

These tests can be interpreted in this situation (after the various arguments have been assigned their values) as: (i) find (and label as e) the episode when Lucy came; (ii) find out whether Lucy was then holding the dessert. The operator COME involves further tests which include various perceptual predicates concerning the motion of Lucy; HOLD is itself such a perceptual predicate.

The procedures may fail in a number of ways: the relevant episode may not be found; Lucy might have been carrying something other than dessert. These failures would be interpretable by the executor, who could try alternative ways of answering the question or organise a reply—including that of "Don't know".

They discuss some of the ways in which syntax might influence the translation program, including control instructions such as 'ACHIEVE', which result from commands. And they outline ways in which factors such as intonation, context and intent would influence the processing.

The next problem was that of exploring of lexicon in more detail, having established the kind of machinery the lexicon would be involved in. Initially they attempted to do this using purely perceptual tests in the lexical entries. But that soon became inadequate and they introduced functional information. Thus a table must have a WORK

TOP which is defined not just as being flat—readily testable perceptually—but also it must be "large enough, and rigid enough, so that, if it is supported horizontally at an appropriate height, it will support objects for a person to manipulate with his hands." Such a definition incorporates sensorimotor predicates in its evaluation and the need for analogic prediction. Consideration of speaker as well as hearer necessitated a more flexible approach to the lexicon, and the need to include in the theory some acknowledgement of the relationships between Lexical items led to the idea of core concepts in semantic fields.

The core of FURNITURE is that it assists in accommodating people's bodies and the objects and instruments they use as they engage in eating, sleeping, working, or playing. This core motivates the conditions in a decision table. The conditions are tests with YES/NO answers and the pattern of answers to the conditions leads to particular lexical concepts. Thus for SEAT there are conditions which ask whether the object (1) is for one person only, (2) has a backrest, (3) is upholstered and (4) has legs. If the first two answers are YES then the object must be a CHAIR; with the pattern YNYN it could be a footstool or an ottoman, and a further test or specification would have to be made. The advantage of the decision tables is that they can be used in both ways—from properties to lexical concepts or *vice versa*. Verbs are related together in a slightly different way in this theory; there are core concepts such as TRAVEL, POSSESS, SEE which are then refined in various ways, such as by specifying the manner—to give verbs such as LURCH, OWN and GLIMPSE.

The semantics is not just of language but of the real world as we perceive it: objects, spatial relations, actions, time; and the use of psychological data is more apparent on the perceptual side of their theory than on the language side. The authors seem to make the Whorfian decision to base perception on language—in this case English. All words are related to the perceptual world and all aspects of the perceptual world are related directly to the lexicon-based conceptual structure. What they do not do is create an intermediate level. The decision tables seem to be language specific, and it is not clear how we can think other than verbally. To take a simple example: I feel I can conceptualise separately: we (me, you and him); we (me and you); and we (me and him). The decision table, however, produces only the conditions—the tests of who is included—and the words. The three versions of *we* are separated only in the columns

of the decision table, which are not the right kinds of things to serve as cognitive units. Their decision may have been the right one but I would have liked to see their motivation.

I expect there are going to be lots of flaws in the routines they suggest. I didn't specially look for them but some jumped out at me—such as the requirement, in the definition of LOOK AT, that the back of the eyes, the pupil and the object looked at have to be in line. Not only can you look at someone through a mirror but also out of the corner of one's eye. The former objection may be trivial but the latter looks as though it will require a distinction to be made between the routines used by the looker and by the looked at. This is a game the authors couldn't hope to win first time out. Either they analyse a few concepts into the ground and produce something lacking generality or they attempt to cover enough ground to justify general principles with the risk of getting details wrong.

While their local arguments are coherent, the book as a mass is rather heavy. I get the feeling that they were looking over their shoulders at linguists and philosophers, and in so doing blurred their psychological position. Their final 'Conclusions' chapter, which might have clarified this, reflects rather the "thank god that's over" feeling of the Preface. One little thing which made the book more difficult to read than it need be is the idiosyncratic indexing. They construct lexical entries for scores of words but don't include the words explicitly in the index. As it is, PERSUADE can be found through 'Communication verbs: perlocutionary'; and core concepts in their model, such as 'core concept', have no entry at all.

To obtain a rounded and more leisurely evaluation of this book (without actually reading it, that is) you will have to watch out for reviews in linguistic, philosophical and psychological journals. Each will have different objectives, requirements and criteria, and together they will pick it over as thoroughly as it deserves. At the moment I have no idea how many of the authors' ideas will survive, but I am certain that it will provide a fresh impetus to work in the area. For whatever its technical weaknesses may be, the outstanding merit of the book is to show how difficult the task is by submersion rather than dipping in the big toe. People really interested in the area have no option but to look at it.

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Biographical springboard

June Goodfield

Half a Century of Medical Research. Vol. 2: The Program of the Medical Research Council, UK. By A. Landsborough Thompson. (HMSO: London, August 1976.) £10.

THIS is the second of two volumes which together cover half a century of research effort, and form a history of Britain's remarkable and distinguished institution—The Medical Research Council (MRC). The first volume (for review, see *Nature*, **252**, 134, 1974) covered the constitutional and organisational aspects of the Council's history and dealt with the intriguing issues of the origins of policy and, "the formulation of those administrative principles" that related to the promotion of medical research. It could have been—should have been—a fascinating book: in the event it is impeccably dull.

This second volume is concerned with the Council's scientific program in detail: once again with the origin of policy and the varied circumstances which led to certain subjects being selected for special support. But now the context is broader for as the MRC itself was expanding over the sixty years in question, so too were the boundaries of the scientific world. So the Council's efforts and achievements must be assessed in terms of the wider international effort in biomedical research.

By any standards the record is most impressive. For measured either by an intuitive feel for fields likely to be significant, or by a balanced stance that recognised the legitimate interests of both the government and the scientific community, serving the one without stifling the other, the record is, on the whole, superb. One can be thankful that, in 1913, the committee that submitted a general scheme of research ignored the narrow interpretation of the phrase—"provision of research"—laid down by considered legal opinion. The National Insurance Act of 1911 appropriated one penny per annum per head of the insured population of Britain, to be allocated from parliamentary funds for "the purpose of research", and the lawyers held that research might well have to be restricted to the diseases of *insured persons* only. At that time, tuberculosis certainly, but possibly not cancer! But the committee quietly by-passed that

issue and we may thank God for that, for stemming from their general injunction to cover "all researches bearing on health and disease, whether or not such researches have any direct . . . bearing on any particular disease", has flowed such specifics as penicillin, the structure of DNA and the basic mechanisms of cell-mediated immunity.

One must accept that this is essentially a work of compilation. By far the greatest source of material used by the author has been the Council's own annual reports and a variety of scientific papers, reports and documents. These are bound to be bland, objective and reasonably free of human virtue, or vice, alike. So as with the first volume, this is not history as historians know it, for the human content has once more been either ignored or extracted. What has been provided is a valuable series of reference points from which others may start more vital histories.

Nevertheless, there are fascinating insights to be discerned for those who are prepared to weave a passage through the maze of detail. The social picture of Britain we have derived from other sources is intensified by a knowledge of where the early clinical efforts were directed—on tuberculosis, rickets, the hygiene regulations for milk—problems that later society does not face, either because of new knowledge or changed habits.

In the days of quack cancer cures it is salutary to be reminded once again of the human traits of venality and fear; much trouble stemmed in the early days from those who promoted dubious claims for tuberculosis. Then, as now, they "stimulated pressure groups of busybodies, and patients,

anxious to pioneer against the alleged tyranny of the professional closed shop". To set this into the contemporary situation, simply substitute "cancer" for "tuberculosis", "lactrile" from apricot pits, for a "powder with a resolute Zulu name extracted from the root of a South African plant", and the "contemporary medical profession together with the US Food and Drug Administration", for a "professional closed shop".

Even Dr Summerlin and the affair of the painted mouse had its counterpart within the Institute's hallowed walls, for in 1927, virology revealed a seamy side. One investigator claimed to have discovered a virus as the causative agent for disseminated sclerosis; she even prepared a vaccine from cultures which was actually clinically used by certain physicians, even before other scientists had tried to repeat the results. When the results could not be reproduced the scientist was invited to repeat her work under supervision. She then "withdrew from the scene", as the author coyly writes, and it was discovered that she had been using the virus of bovine pleuropneumonia which had been described by the National Institute, and from where she had obtained her cultures.

Plus ça change . . . but things are never quite the same. Science in its achievements has progressed wonderfully even if human beings seem to be much the same. But they are deeply fascinating nevertheless, and I miss them in this volume, just as much as I did in the first. □

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Fact, fiction and fraud

Michael Stoker

The Patchwork Mouse: Politics and Intrigue in the Campaign to Conquer Cancer. By Joseph Hixson. Pp. x+228. (Anchor/Doubleday: Garden City, New York, 1976). \$7.95.

IN this book Joseph Hixson tells the story of Dr William Summerlin, and much more besides. Summerlin, a dermatologist turned immunologist, claimed that tissue which had been kept in culture could be grafted across transplantation, even species, barriers. But, in 1974, when working at the Sloan-Kettering Institute in New York, he



William Summerlin

was caught red (or rather black) handed after using a felt pen to touch up some supposedly successful pigmented skin grafts in white mice. The subsequent enquiry revealed other evidence or de-



Robert Good

liberate misrepresentation to his colleagues. For example, untouched rabbit's eyes had been shown at several meetings as successful corneal transplants.

A nasty little episode no doubt, and pretty awful for those involved, but why should a distinguished science writer bother to write a book about it? After all it is not the first time that a scientist has been caught cheating; and, paradoxically, where fiddling of evidence is the greatest sin, as in scientific research, it also has the least consequence in the long run, since the cheat destroys himself, not science.

The Summerlin incident had special features, however, which must have proved irresistible to Hixson, and which he has exploited with skill and flair. First, the claim itself was of tremendous potential importance, not only for patients waiting for grafts, but for its bearing on immunological theory, based as it is on the classical concepts of Burnet and Medawar. All this is painlessly explained in considerable detail and with the expertise of the good professional journalist. Second, Summerlin, not normally one for rushing into print with detailed articles in scientific journals, is apparently a charming and persuasive speaker, and his transplantation story had received the full treatment from the press and radio networks. Small wonder that the media were not silent when the science writers found they had been caught out. Hixson's often riotous, but always sympathetic, accounts of his colleagues in the press corps are not to be missed, if only for the educational content, which should be recommended reading for graduate students.

Most important, however, in Hixson's view, was Summerlin's relationship with Dr Robert Good, Director of the Sloan-Kettering Research Institute, the largest cancer research establishment in the United States, one of the original triumvirate advising the

President on the great cancer programme and one of the most powerful figures in immunology and cancer research. Good had been intrigued by Summerlin's claims, and brought him to New York in an exalted position, apparently to the disquiet of some of the original senior staff of the Institute, who were inevitably grumbling about a new Director riding roughshod over the existing regime, and pushing on his own ideas, especially on tumour immunology, to the forefront. Good, although a co-author with Summerlin (presumably as his sponsor) had never been directly involved in the work himself, and partly as a result of reports from Summerlin's own staff, had indeed begun to suspect something fishy some months before the ink episode. Summerlin's defence was that he broke down in the face of Good's insatiable demand for significant data (they were certainly needed); and instead of being summarily sacked, as many might believe should follow such dishonesty, he was actually rewarded with one year of terminal sick leave.

The pressures in the Sloan-Kettering, and its special position, thus leads Hixson inevitably to the uneasy power politics of the proposed (and ordained) 'Conquest of Cancer'. Although it is all dealt with historically, and with little explicit statement of the author's personal view, the message which emerges in Hixson's book is clear—that vote-seeking congressmen, anxious administrators and a hungry press, are tempting greedy scientists into exaggerated predictions and claims about cancer, which if not actually as fraudulent as those of Summerlin, may seem so in retrospect to relatives of cancer patients.

The Patchwork Mouse is not written for the usual *Nature* reader, and you have to swallow a colourful narrative style, which may not always be based on direct observation. "Robert Good has shaved and pulled a white turtle neck shirt over his dark, tousled head at an hour when the swinging East Side singles bars have barely closed their doors". How does he know? Perhaps Good gets into his turtle neck feet first. There are also a few errors. For example, cancer as a whole is not commoner in those with defective immunity, only certain rare forms of the disease. The text seems, however, to be generally accurate with careful qualification of statements and quotations whenever required.

This is a fascinating and readable story with compassion for the people involved. Unlike the principal character, Hixson tells it as it is. □

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Memorial to cotton research

H. S. Darling

Agricultural Research for Development: The Namulonge Contribution. Edited by M. H. Arnold. Pp. x+353. (Cambridge University: Cambridge, London and New York, August 1976.) £13.

THE Cotton Research Corporation (CRC) was established by Royal Charter in 1921 under its former title of the Empire Cotton-Growing Corporation. For 55 years until it was dissolved in 1976, the CRC played a leading role in research and development in support of the field production of the cotton crop in the anglophone countries of the tropics especially in Africa. From its administrative headquarters in London the CRC maintained a team of thirty to forty carefully selected and trained research workers distributed around the world and renowned alike for their unity of purpose and the consistent high quality of their work. The CRC team was regarded with considerable justification as a *corps d'élite* among the British agricultural scientists who have made such an important contribution to the development of the anglophone territories of the Third World.

The deployment of CRC staff was governed from the outset by two considerations. The first was the need to provide research assistance, often of a trouble-shooting nature, in areas in which increased cotton production was desired. In such cases, CRC workers were posted to government agricultural stations in cotton-growing areas to give specialist support to local research programmes. The second consideration was the need to maintain at least one permanent multidisciplinary research centre under direct CRC control, where long-term work of a fundamental nature could be carried out on problems peculiar to the cotton crop with the object of providing information of relevance throughout the tropics. From 1945 to 1972 this second need was met by the Namulonge Cotton Research Station in Uganda.

As its subtitle indicates, the book under review is concerned with Namulonge and is a composite production from the pens of senior scientists, each a specialist in his own field, formerly on the staff there. Editorial unity has been gently imposed by Dr M. H.

Arnold, the last CRC Director of the station, who is himself a major contributor. The whole provides an excellent description of Namulonge and of its work over the period 1945-72. Sited in the centre of the southern half of Uganda some 35 km north of Kampala, the station covered 900 hectares of reasonably fertile soil, most of which was suitable for field trials and other forms of agricultural research. Established on land leased by the CRC from a Ugandan landlord and built with UK funds, the station was well equipped with laboratories, other research facilities and housing for staff. In 1972 the station was handed over to the Ugandan Government as a going concern some three years before the CRC itself came to the end of its active life.

The meat of the book lies in carefully written yet readable descriptions of research at Namulonge given in chapters 2-8 and comprising two-thirds of the total text. These seven chapters bear the following titles: agrometeorology; soil productivity; crop physiology; entomology; plant pathology; resistance breeding; and plant breeding. They are followed by a chapter of importance on Namulonge farm and a closing chapter on the application of agricultural science to national development. There is also a references and author index and a full subject index.

The accounts of research are always competent and often brilliant. The descriptions of cotton-breeding in chapters 7 and 8 are highly authoritative, embodying as they do cumulative experience of 40 years of work by several generations of CRC geneticists and breeders including men of the standing of J. B. Hutchinson and R. L. Knight. Surprisingly enough, the name of S. C. Harland is conspicuous by its absence! The chapter on Entomology is to be commended for its clear and accurate analyses of complex situations resulting from the sequential incidence of a wide range of pests, and from the interaction of the effects of insect-damage and planting times. Less satisfactory is the chapter on soils and soil productivity where no mention is made of work in Nigeria by B. W. Bache, P. R. Goldsworthy, R. G. Heathcote, D. Lawes and others, which presumably was well known to the Namulonge team. This West African research preceded and paved the way for the agronomic advances described by the Ugandan workers in the late 1960s and early 1970s.

For all its general scientific competence and its areas of technical brilliance this book is less than satisfactory in that it fails to ask, let alone seek to answer, at least three important questions. First: why was Namulonge

chosen as a site for basic research on cotton-growing? Second: why does no thought seem to have been given in the planning of work programs to research on the human factors that dominate cotton production? Third: why is so little mention made of plans for training local research workers to replace European staff when (as has already happened) CRC control came to an end? These questions are of major significance.

The choice of Namulonge was made when the reviewer was working as an agricultural research worker in the then Ugandan Protectorate Department of Agriculture. It seemed obvious that the site was atypical of areas suited to cotton production in that it was too cold and was unsatisfactory in respect of rainfall distribution. It has long since been accepted by Ugandan farmers that the area is agronomically suited to the growth of robusta coffee and plantains rather than cotton. The farming systems laboriously worked out by the Namulonge staff, although successful in themselves for making the best of cotton-growing in local conditions, are largely irrelevant to local agriculture and are to that extent a waste of effort.

Successful cotton-growing in the near-subsistence conditions that obtain in Ugandan agriculture, and indeed throughout much of tropical Africa, depends on plentiful supplies of human labour. Any research program aimed at increasing cotton production in developing countries should recognise the importance of human factors and, where these factors are not clearly understood, should seek to define and elucidate them. Technical developments conceived without relevance to human considerations often fail to be adopted. A classic example of this is the question of planting dates for cotton in Uganda. Here scientific evidence indicated that early planting was essential for best results in terms of yield. All human experience showed that local farmers found such early planting to be impracticable in the context of their overall farming systems. In the view of the British research worker, trained in the natural sciences, it was essential that early planting be advocated as official policy and the resulting impasse has dominated local cotton-growing policy for many years!

Namulonge, in common with many other British-based agricultural research stations, failed to appreciate the need for research inputs from economists and rural sociologists in the interdisciplinary mix of the station program as a whole. The failure is apparent in the last chapter of the book on the application of agricultural science to national development, which is too vague to attract attention or

inspire confidence. The workers at Namulonge clearly felt confident of their ability to advise on growing cotton. It is equally clear that they had few ideas as to the role of cotton production in the Ugandan economy.

The apparent lack of attention to the training of African staff is surprising. The upgrading in 1948 of Makerere College at Kampala to university status enjoying special relationship with London University followed closely on the establishment of Namulonge in 1945. The first director at Namulonge, Sir Joseph Hutchinson, was Chairman of the Council of Makerere College from 1953 to 1957, and played an important role in the development of the excellent Faculty of Agriculture there, including the establishment of its outstation at Kabanyolo. Namulonge's staff were

encouraged to lecture in the Faculty. Yet in spite of these close ties between Namulonge and Makerere, little or no thought seems to have been given to using the new university to build up a body of local scientific expertise in cotton research.

This grave omission was unfortunately a major weakness of the CRC as a whole. The dissolution of the Corporation in 1976 has consequently meant that with the disappearance of the London headquarters, little in the way of research staff and human assets persist in a recognisable form. A proud ship has sunk virtually without trace and the book under review can almost be regarded as its memorial tablet. □

Dr Darling is Principal of Wye College, University of London, UK.

Self-reliant development

C. H. G. Oldham

Beginnings of Brazilian Science: Oswaldo Cruz, Medical Research and Policy, 1890-1920. By Nancy Stepan. Pp. xi+225. (Science History: New York, June 1976.) \$12.95.

EFFORTS to build up local research capabilities in many developing countries over the past two decades have frequently been frustrated by the lack of local demand for the results of this research. As a result, policy makers in these countries are currently preoccupied with measures to stimulate demand, and to make science more relevant to national needs. This is proving to be no easy task since the science system has grown in isolation from the productive system and the forging of links is exceedingly difficult.

Nancy Stepan's excellent book, *The Beginnings of Brazilian Science*, shows how governments in the developing world might have been saved a good deal of unnecessary expense and anguish had they known of, and heeded the lessons which were to be learnt from the growth of biomedical science in the late nineteenth and early twentieth centuries in Brazil.

Most of Professor Stepan's book, and certainly the best part of it, is devoted to a case study of the Oswaldo Cruz Institute in Rio de Janeiro. This Institute was initially established in 1900 to produce yellow fever serum but grew into an excellent scientific centre covering the entire spectrum of scien-



tific activities—basic research, development, staff training and even production. It had an international reputation and was staffed entirely by Brazilian scientists. Those looking for an example of self-reliant development in science in the third world would have to search hard to find a better example than the Oswaldo Cruz Institute. To emphasise the positive lessons which she thinks are to be learnt from the history of this Institute, Professor Stepan contrasts its success with the failure of the Bacteriological Institute of Sao Paulo, established at roughly the same time. The story of the Sao Paulo Institute, although useful for comparative purposes, palls in comparison with the exciting and dramatic success of its Rio de Janeiro rival.

Professor Stepan clearly shows that the success of the Oswaldo Cruz Institute owes a great deal to the happy coincidence of the right people being brought together at the right time and the right place to meet a particular need. The need was particularly urgent. Brazil, at the turn of the century, had

most of the physical ingredients required for the development of a prosperous and wealthy nation. The main drawback was its extremely unhealthy climate. Rio de Janeiro was largely avoided by foreign visitors because of its foul reputation for disease. Epidemics of yellow fever, bubonic plague and other tropical diseases caused the deaths of thousands of people each year. The need for a solution to these epidemics was apparent to the public and politicians alike. There was a 'demand' for the results of scientific research.

Fortunately the time was also ripe scientifically for a biomedical research institute established in a developing country to be able to make internationally significant contributions, and at the same time solve a pressing national need. Had the politicians provided the funds thirty years earlier, it would have been too soon. But, in the latter part of the nineteenth century, due in large measure to the work of the Pasteur Institute in Paris, an understanding of the bacteriological origins of many diseases had been developed and the whole field of immunology had been opened up. By 1900, when the Institute was founded, there was a real opportunity for major advances to be made in bacteriology; an opportunity which the Brazilian scientists were not slow to exploit not only for the benefit of the Brazilian people, but also for the benefit of science as a whole.

The real hero of the story is Oswaldo Cruz himself. He was trained as a doctor and went to study at the Pasteur Institute in Paris. He returned to Brazil and, against very considerable political and public opposition, converted an Institute initially established solely to manufacture serum, into one of the world's leading biomedical centres. His approach was almost the exact opposite of the one normally taken by developing countries today. Instead of starting with research and then trying to find some use for the results, Cruz began with a need, and with a solution to a part of that need—the manufacture of yellow fever serum. He then worked from this to applied research and eventually basic research. This distinction between basic and applied research, however, does not seem to have been one which bothered those Brazilian scientists working at the Oswaldo Cruz Institute. For them there was a job to be done, and if this job required advancing the frontiers of science, this was part and parcel of the job. If on the other hand all that was required was some simple adaptive work in development, then this too was carried out. The Sao Paulo Institute never developed the same range of scientific

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activities and, according to Nancy Stepan's analysis, suffered as a consequence.

The book is a fascinating story of the interplay between science, politics and development as they relate to the growth and utilisation of the biomedical sciences in Brazil in the first three decades of the twentieth century. It is marred only by an over-ambitious title (the book only treats some aspects of the biomedical sciences, and not, as it implies, science as a whole), and by an over-simplistic final chapter on the science policy problems of developing countries. There are lessons to be learned from the experience of successful and unsuccessful institutions, and Professor Stepan is right in suggesting

that some of the lessons to be learned from the Oswaldo Cruz Institute are as valid today as they were in the early twentieth century. The times, international relationships, and local political situations have all changed, and Nancy Stepan probably exaggerates the extent to which the lessons have relevance for policy makers today. Nevertheless it is a good book, well researched, and is highly recommended to anyone interested in the broader policy implications of biomedical research, public health, Brazil, and development. □

Dr Oldham is Deputy Director of the Science Policy Research Unit, University of Sussex, UK.

Horological insights

A. A. Treherne

The Lost Science of John "Longitude" Harrison. By W. S. Laycock. Pp.159. (Brant: Ashford, Kent, 1976.) £18.75; \$45.75.

It is fitting that in this year, which marks the bicentenary of John Harrison's death, a book should be published which establishes clearly his contribution to science, and in particular to the science of horology. Even if the reader cannot follow the author in believing that Harrison's contributions to the science (as distinct from the practice) of horology surpass those of even Huygens, the author does establish beyond reasonable doubt that Harrison deserves more than the standard, cursory mention in the physics textbooks for his grid-iron pendulum—a remarkable achievement in itself which is, perhaps, for the first time adequately treated by the present author.

Harrison's contemporaries could never quite bring themselves to believe that a man without the standard academic credentials or any formal horological training could make significant contributions to horological science. Hence the reluctance of the Board of Longitude to award to Harrison the full £20,000 prize offered by the British Government for a solution to the problem of determining longitude at sea, even though during the sea trial of 1761 his 'sea clock' performed well within the limits laid down. Harrison's unorthodox ideas seemed perverse or even absurd in the light of the established wisdom of his time and in spite of his practical successes few made any attempt to understand the principles behind his timekeepers

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ignoring his published writings as the polemical abuse of a frustrated man. Later historians of science have even less excuse than Harrison's contemporaries for not recognising his successes as arising from the application of scientific method and principles, and for not seeing his work as a clear and natural continuation of the work of Huygens. It is refreshing to find the case so well argued in this book.

The unconventional methods which Harrison adopts are convincingly analysed and justified by the author, again perhaps for the first time. The interdependence of his ideas, and the important role of his remarkable achievement in completely eliminating the need for lubricants, emerge especially clearly; underlining the impossibility of making sense of these ideas if they are considered in isolation, as they often have been in the past. In fact the author not only argues that there are good scientific grounds for Harrison's apparently fantastic claim that it would be possible to make a clock according to his principles with a rate of 2 or 3 s yr⁻¹, he also outlines a programme, already begun, to construct a pair of regulators based on Harrison's ideas, in order to put the claim to the test.

Harrison's writings are by no means easy to interpret, as is witnessed by the

fact that no convincing interpretation had previously been offered. What we have in this book is an ingenious but coherent interpretation which, in spite of the necessity for a good deal of imaginative reconstruction and speculation, is, in broad terms, convincing. Moreover, the analysis is always stimulating and could hardly fail to reveal many new insights. I have two main regrets. First, the exorbitant price of the book. At this price one might have expected facsimile reproductions or transcriptions of Harrison's texts as well, which would largely have nullified my second concern: that it is not always easy or even possible to tell from the book alone what is in Harrison's texts and what is speculation or

reconstruction. There are some mistakes and misprints, which most readers will have no trouble in spotting and correcting, but which would prove confusing to a reader with little scientific background.

This book is an important contribution to horology and to the history of science. The general reader, who will require only a very elementary background in mathematics and physics to follow the arguments, will also find it fascinating and stimulating. If it does not sell well, it will be because of the price rather than the content. □

Alan Treherne is a Lecturer in Mathematics and Philosophy at the University of Keele, UK.

Ecology of plankton

G. E. Fogg

Marine Plankton Ecology. By Paul Bougis. Pp. ix+355. North Holland: Amsterdam and Oxford; American Elsevier: New York, 1976.) Dfl.130; \$49.95.

In some ways the ecology of plankton may be simpler than that of terrestrial communities. For most purposes there is only one phase to deal with and the organisms are generally smaller and less complicated than those on land. But apart from the inconvenience, expense and limitations of working at sea, the marine plankton ecologist has the difficulties of investigating events, following each other much more rapidly than in terrestrial situations and in an ever-shifting medium, which may be directly related to others that have taken place thousands of miles away. It is only comparatively recently that the resources needed to deal, even inadequately, with this situation have become available and the writers of textbooks are only just beginning to catch up.

Professor Bougis, who is Director of the Zoological Station at Villefranche sur Mer, has, however, produced an admirable and comprehensive account of this rapidly developing subject. He deals with phytoplankton and zooplankton equally, and with each he begins with a necessarily brief survey of the systematics of the amazing variety of organisms concerned.

For the phytoplankton he devotes chapters to the factors affecting photosynthesis, the nitrogen and phosphorus cycles, other nutrient elements and organic growth factors, enumeration and periodicity, and primary produc-

tivity. For the zooplankton he gives accounts of quantitative studies and distribution, vertical distribution and diurnal migration, nutrition, metabolism and energy conversion, and secondary production.

A final chapter considers the place of plankton in the marine ecosystem, touching on its economic importance and possible exploitation. Appendices deal with such diverse topics as the derivation of the Sverdrup equation defining the conditions required for the spring outburst of phytoplankton and the design and construction of plankton nets—information which is not readily obtainable from other textbooks.

With such a broad canvas it is inevitable that some areas should not be as well covered as others and one such is the topic of phytoplankton buoyancy, which is treated simply as a matter of cells remaining afloat in the illuminated surface waters. The possible value of sinking as a means of increasing nutrient uptake, its interaction with wind-induced Langmuir circulation (which is mentioned in relation to zooplankton distribution only), and the possible role of differential rates of sedimentation in determining species succession, are not considered.

The book is well illustrated and the translator has done an excellent job. There is, however, the increasingly common mistake about the plural of flagellum; fluorescence and luminescence are confused; and a unicellular film of ferric hydroxide is referred to when, presumably, a monomolecular film was in mind. There are also quite a few errors that have escaped the proof-reader. Nevertheless, these are minor matters and the book will undoubtedly be most useful to students, teachers and research workers in marine biology who have access to a library that can afford to buy it. □

G. E. Fogg, FRS, is Professor of Marine Biology at University College, North Wales, Bangor, UK.

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University of Odense, Denmark

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H SMITH

University of Nottingham

This volume provides a fully comprehensive and up-to-date summary of the control by light of the growth and development of higher plants. It presents the proceedings of the 22nd Nottingham Easter School in Agricultural Science, at which specialists from science, commerce, industry and practical agriculture discussed the important question of Light and Plant Development.

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Too little, too late

H. I. S. Thirlaway

Nuclear Explosions and Earthquakes: The Parted Veil. By Bruce A. Bolt. Pp. xxi+309. (Freeman: San Francisco and Reading, March 1976.) £6.

AFTER the Soviets had unexpectedly broken the moratorium on nuclear tests in 1961, the distinguished owner of this journal (Harold Macmillan) was said to have asked his advisors to show cause why his Government should continue to support seismological research for a test ban; after all was not the UK also free of serious earthquakes? Happily the advice was convincing, the work continued and, according to Professor Bolt, to some effect in a country which had lost its impetus for experimental seismology after John Milne and his contemporaries.

I was reminded of this anecdote by the author's surprise that seismology in aseismic Sweden should be so well supported. It is good that it is because, like the Shields which are seismologically exploited by the Commonwealth countries, Sweden is as near perfect a receptor for seismic waves as one could hope to find—Uppsala was well known for its overestimates of the seismic yields of underground explosions—and in spite of such sites the technical reason why there is no ban on nuclear tests is because the threshold of seismic detection is still rather too high to deter cheating. The fact that places where the seismic transmission properties and ambient noise might be good enough for seismic monitoring of a comprehensive ban on tests are unlikely to be found is not made too clear by Professor Bolt. This means that as long as the possibility of secret tests is a cause for concern, previously unacceptable threshold treaties, like that of the new Soviet-American agreement, must suffice. The author concludes: "the veil across the future of history of civilisation has not been parted".

Nonetheless, seismologists were given every encouragement and opportunity to make a significant contribution to the unveiling; and in describing how they failed this new book by Professor Bolt gives the lay reader a broad and well balanced insight on a many-sided problem.

The basic mechanisms and effects of nuclear explosions and earthquakes, the

way in which the Earth's interior structure and the seismograph modifies the relatively simple seismic wave radiations from these sources, are explained in the first four chapters. We are given an inkling of the confusion which is to follow, but at first all seemed straightforward to the geophysicists from East and West who gathered in Geneva in the summer of 1958 to examine the possibility of monitoring compliance with a treaty to ban nuclear tests. Ironically, the weapon tests which were to deny some of their predictions had already been planned for the autumn; only a month or two after the publication of their report describing how 5 kton or more could be reliably monitored, American seismologists experimentally demonstrated that there would be a great deal of doubt about underground explosions of as much as 20 kton and a few months after that, American physicists showed (theoretically) how to convert a megaton to a few seismic kilotons by detonating in a large enough hole in the ground. That it had to be an unbelievably large hole was irrelevant; the damage was done, in the next three years the opportunity to change the course of affairs (whether for good is another question) passed and the ensuing decade has been devoted to the founding of forensic seismology.

In the end it has turned out that the 1958 experts were not so far wrong, on average, in their 'guesstimate' of 5 kton; seismic waves from yields of this order (as reported by the Soviets) have been recorded on western seismographs with disarming clarity. The trouble is that signals from Nevada are not so well received and it is on Nevada Test Site experience that the Earth is calibrated in terms of seismic magnitude to kilotons of explosive. Geophysicists have been slow to accept that the property of the upper mantle to absorb seismic wave energy varies widely according to the tectonic history of the region. For policing a test ban, it is the behaviour of explosions in regions of high attenuation which determines the threshold at which compliance with a test ban can be guaranteed.

Together with an overestimate in the number of earthquakes of a given size to be expected each year (an error which was rectified in a little remembered US Atomic Energy Commission announcement in July 1962), the existence of unknown, highly attenuating structures was the fundamental source of the early confusion. Professor Bolt takes the next three chapters to describe the results, in the course of which he outlines the contributions of the USA, the Soviet Union and the UK, and of Australia, Canada, China,

France, Japan and Sweden. The Berkner Panel Report outlined the needs in the USA and a curiosity of this report is that its principal contributor was John Gerrard, a product of Professor Bullard's Cambridge department which was to play a useful role in the UK's own contribution.

At the time Bolt completed his book, the prime importance of lateral variations in the upper mantle had not been established, nor had seismogram modelling and the refined aspects of broad band seismology been confirmed as techniques for discriminating between seismic sources and for revealing some methods for testing in secrecy; it says much for the still vigorous development of forensic seismology that Professor Bolt's book is already out of date.

The last three chapters are devoted to accounts of spin-off from the development of nuclear weapons. These include: nuclear engineering for which Professor Bolt is an enthusiast in the face of counter arguments that it is not realistic to seek test bans and non-proliferation of nuclear weapons with, at the same time, freedom to engineer with nuclear explosions; the environmental consequences on people, animals and on earthquakes, the controversies about which were initiated by the megaton sized tests in Nevada and Amchitka Island and, finally, the discoveries about the interior of the Earth which have been made possible because the hypocentre, origin time and size (all unknown quantities for earthquakes) can be measured with great accuracy for nuclear explosions. These discoveries are grouped in the chapter on seismology and nuclear politics on the dubious grounds that politics prevents the free exploitation of large explosions in the interests of seismology.

The book is equipped with appendices giving incomplete lists of nuclear tests by the USA, the Soviet Union, the UK, France and China, and the full text of the 1963 Test Ban Treaty. It is prefaced by a helpful chronology on the principal nuclear and seismological events beginning with Trinity (the first test of all) and ending with the first underground test by France in the south-west Pacific. To students of the test ban problem this French test may represent another gain for the 1963 treaty in spite of non-adherence to it by France. Professor Bolt uses boxes to enclose the more technical sidelines and thereby neatly maintains the well written flow of the text without interruption. □

Dr Thirlaway is Head of the Seismological Research Group at the Ministry of Defence Atomic Weapons Research Establishment, Blacknest, UK.

Oil shale

R. C. Selley

Oil Shale. (Developments in Petroleum Science, 5.) Edited by Teh Fu Yen and George V. Chilingarian. Pp. xi+292. (Elsevier Scientific: Amsterdam, Oxford and New York, 1976.) Dfl.90; \$34.75.

OIL SHALE is a fine-grained organic-rich sedimentary rock which releases crude oil when heated. The industrial extraction of oil from shale began in Scotland in 1862, but at the present time the oil shale industry is moribund in the UK. (A review of British oil shales has been given in a recent paper published by the Department of Energy.)

Over much of the Western World the oil shale industry was important in the past, and will be so in the future, but is dormant at the present time. It is currently practised, however, in the USSR and China. One is reminded of the White Queen's rule: 'Jam tomorrow and jam yesterday-but never jam today' (Carroll, *Alice through the Looking Glass*, Macmillan, 1871).

The world's resources of oil shale may contain some 30×10^{18} barrels of oil, only 2% of which is available for present-day commercial exploitation. This figure is controlled by two factors: the price of crude oil and extraction technology.

The conventional method of extraction necessitates mining the shale and distilling off its oil by retorting above ground. Subsurface *in situ* retorting, aided by explosive fracturing and fluid injection, is still in the experimental stage. So too are biochemical extraction methods currently under investigation at the University of California.

Due to recent advances in organic geochemistry the composition and molecular structure of oil shale hydrocarbons are well known. Similarly the geographical distribution and the geological factors which control the formation of oil shales are well documented. All that is needed to revive the oil shale industry is either a further increase in the price of crude oil, or a major breakthrough in the technology of extracting oil from oil shales.

It is a cliché of our time to say that oil is too precious to burn. It should be used only as the foundation of the petrochemical industry, never as an energy resource. Nonetheless, if nuclear, geothermal or any of the other energy sources currently under review fail to supply the demand then oil shales may become very important.

Oil Shale provides a useful review

of current knowledge of many aspects of oil shales. The book consists of twelve chapters edited by T. F. Yen, Associate Professor in the Department of Chemical Engineering in the University of Southern California, and G. V. Chilingarian, Professor, His Imperial Majesty Shahanshah Arya Mehr Chair of Petroleum Engineering, in the same university.

It is true to say that the University of Southern California is the intellectual font of the book, providing a third of the sixteen contributing authors. The geological core of the book is undoubtedly the Green River oil shale, which, according to the index, is mentioned on 66 pages of the 266 pages of text.

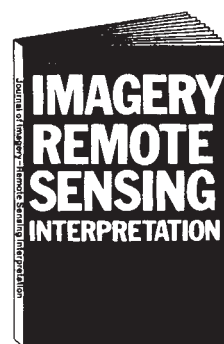
Nonetheless, the book covers the field well. It begins with a brief review of the topic. This is followed by an account of the geological setting of oil shales and their geographical distribution. The latter is displayed in a series of maps showing the distribution of oil shales during different geological periods. This could have been greatly improved had the data also been tabulated so that the reader could find the literature source for every oil shale located on the maps.

An account of the origin and formation of oil shale follows, and is succeeded by a chapter on the origin and characteristics of the Green River oil shale. This Tertiary lagoonal deposit covers large areas of the states of Colorado, Utah and Wyoming, and contains some 80×10^{12} barrels of oil (recoverable by existing technology), representing 15 years reserves of crude for the USA at the 1973 consumption level.

The following chapter, on the mineralogy of oil shales, in point of fact contains a fuller account of the geology, as well as the composition, of the Green River oil shales, than that which precedes it. Succeeding chapters review retorting technology, well-logging techniques, environmental implications (of course) and the history of oil shale research over the past 30 years.

Considered overall, therefore, this book provides an important and comprehensive corpus of up-to-date information on the geology, chemistry and extractive technology of oil shale, which will be of interest to geologists, chemical engineers, government officials and other assorted doom-watchers. In common with most books from this publisher its price of about £20 (at today's rate) puts it beyond the reach of everyone except pop stars and oil sheiks. □

Dr Selley is a Reader in Petroleum Geology at Imperial College, University of London, UK.



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Friendly persuasion on the energy crisis

John Chesshire

The Poverty of Power: Energy and the Economic Crisis. By Barry Commoner. Pp. 314. (Knopf: New York, 1976.) \$10.

ALTHOUGH the world may be experiencing signs of an imminent or longer term shortage of energy, there is certainly no shortage of those prepared to write about it. This makes Barry Commoner's latest book all the more welcome because of its refreshingly different approach to energy problems. Although by training a biologist, Commoner uses the physicist's tools (the First and Second Laws of Thermodynamics and the concept of entropy) in a fluent manner to reach the apparently simple, yet powerful, conclusion that "energy is efficiently used when the quality of the source is matched to the quality demanded by the task".

The book's coverage is restricted to the USA, which accounts for over 30% of world commercial energy use, and comprises chapters on the major fossil fuels, nuclear power, solar energy sources, the energy demand sectors and the increasing capital intensity of additional conventional energy supplies.

Although the First Law of Thermodynamics has long been used to assess the efficiency of energy use, it is only since the quintupling of oil prices after October 1973 that energy studies using the Second Law have been widely undertaken. Such studies have revealed that in many instances efficiencies measured according to the precepts of the Second Law are ten times lower than the better-known First Law efficiencies. For example, the First Law efficiency of typical oil-based domestic central heating is 60%, whereas its Second Law efficiency is only about 8%. The installation of a heat-pump system, however, can raise this above 20%.

In his examination of the conflicting evidence on world fossil fuel reserves Commoner is highly sceptical of the value of estimates at present widely agreed. In the case of assessments of US crude oil reserves Commoner argues that "the declining rate of oil discovery per year is a result of company decisions to cut back on exploration efforts rather than on the

depletion of accessible oil deposits". The low economic returns from exploration in the USA have led to the increasing multinationalism of the US petroleum majors seeking lower cost (and higher profit) sources in the Middle East and elsewhere. Nevertheless, despite his optimism regarding the potential size of US and world reserves, Commoner argues that the physical fact of exhaustion, even if delayed by potentially greater resources, is reason enough to plan for the transition from the present heavy dependence on oil to renewable energy sources.

Although Commoner systematically reviews many of these benign energy sources he seems most committed in his discussion of the many issues raised by the growing utilisation of nuclear power. The Atoms for Peace program launched by Eisenhower in 1953 ushered in the widespread harnessing of nuclear power for civilian purposes, but its earlier, war-time history inevitably resulted in the nuclear power industry being a potentially explosive mixture of the civilian and military.

The economic structure of the US nuclear industry is a complex web of public and private enterprise: research and development are largely government funded; uranium mining, milling and conversion are privately funded; fuel enrichment is carried out in government-owned plants, fuel fabrication privately; commercial reactors are owned and operated by private utilities with substantial government supervision (for example, licensing and inspection), whereas all the proposed waste disposal schemes will be government controlled. In addition, since nuclear power is the only civilian technology that the insurance market is unwilling to underwrite, the Price-Anderson Act ensures substantial state settlements in the event of claims arising from a nuclear accident.

Commoner's concern with the specific technological difficulties and dangers associated with fast breeder reactors and the apparent lack of progress to date on operational long-term waste storage and disposal facilities—"a sign both of commendable caution and inadequate work"—is all the more topical given the recent Royal Commission Report on *Nuclear Power and the Environment*. Although confessing his unfamiliarity with economics, he presents a strong case based on the differential paths of increases in real capital costs and fossil fuel prices, to reveal "the likelihood that the entire nuclear program is headed for extinction". I am not totally convinced by this argument, but it is certainly the case that faced with increasingly politicised debate and

opposition to nuclear power—namely, the Swedish election and the Californian referendum—competing claims on restricted public expenditure and the energy research and development budget, rising capital costs, greater environmental safeguards, and downturns in traditionally exponential electricity growth rates, proponents of nuclear power are facing a major re-assessment of its future role.

In conclusion, Commoner is a persuasive commentator and despite

his strong environmentalist bias and support for solar energy, agrees that the so-called energy gap will not be "swept away in a flood of sunlight". One minor complaint: for a book which is inevitably quantitative, there are no tables or charts to ease the exposition. □

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Energy supply and demand

Peter Chapman

Potential Energy: An Analysis of World Energy Technology. By Michael Kenward. Pp. x+227. (Cambridge University: Cambridge, London and New York, September 1976.) Hardcover £6.20; papercover £2.80.

ENERGY research and development is one of the few fields of inquiry that enjoys a steady growth in its level of financing, perhaps more so in the USA than in the UK. Over the past two and a half years Mike Kenward has charted the ups and downs, the fashions and flops in energy research and development in the bi-weekly 'Energy File' in *New Scientist*. He has now put together a large part of this material in *Potential Energy* which claims to be an "analysis of world energy technology". The review of energy supply options is certainly comprehensive, from solar to nuclear fusion through oil, coal, gas, fission, wind and waves. For anyone not familiar with the range of developments in these fields this book is certainly a useful starting point, but only a starting point.

There are two major defects in this book. The first is that Kenward studiously avoids trying to assess the relative merits and disadvantages of the energy technologies discussed. Occasionally we catch a glimpse of what the author's opinions are: "as far as nuclear research and development goes it is doubtful if even more spending is justified". But by and large he merely describes the technology and describes other people's reactions to the problems. This journalistic style was appropriate for a magazine feature on energy, but leaves the reader of the book in a policy vacuum. This is characterised by the platitudinous and obvious conclusion to the book which points to the twin dangers of "putting all our eggs in one basket only to discover there is a hole in the bottom"

and spreading research and development funds "so widely that no energy technology receives enough support to prove itself". Since this was, and remains, the obvious problem in formulating an energy research and development policy it seems a shame that nowhere does Kenward address himself to the problem.

A more glaring defect in Kenward's treatment of energy research and development policy is the other half of the energy question, namely energy demand. In 218 pages of text there are no more than 8 pages on factors likely to influence energy demand. This reflects the institutional blindspot to the one energy research and development area likely to pay off most handsomely in the medium term. It seems likely that with a moderate range of incentives UK energy demand could be con-

strained to increase by less than 30% over the next 25 years (compared with an increase of 60% over the past 25 years). If this potential exists with present energy utilisation technology it is likely that further research and development on energy utilisation technologies could lead to even greater savings. The known savings are cheaper by a factor of between three and ten, in both capital and total cost terms, than the equivalent increases in energy supplies. This indicates that there is considerable scope for developing additional energy-saving technologies before they cost as much as the energy supply technologies. This is recognised in some of the small print in Walter Marshall's discussion document *Energy R & D in the UK* (Department of Energy, Series of Papers on Energy, June 1976), but is ignored in practice. It seems that Kenward has been unduly influenced by the massive volume of literature and finance associated with energy supply technology and has missed perhaps the largest 'potential energy' area in an otherwise useful review. □

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Climate and landforms

Robert P. Beckinsale

Geomorphology and Climate. Edited by Edward Derbyshire. Pp. xii+512. (Wiley-Interscience: New York and London, August 1976.) £14.50; \$29.

THROUGHOUT the twentieth century the direct and indirect influence of climate on the shapes of landforms has been generally admitted, and the cult of distinctive landforms in distinctive climates has become a hallowed geographical tradition. There is much in its favour because the characteristic landforms in cold glaciated lands, hot deserts and humid tropical regions seem to differ markedly from each other. So climatic geomorphology grew up as a zonal concept wherein each broad thermal belt tended to develop its own specific complex of dynamic processes. Climate was exalted as the great panjandrum, the universal supplier of solar energy and the creator of kinetic energy for the moisture machine. Climatic geomorphologists seemed to be over-riding the doctrine of James Hutton:

"Our land has two extremities; the tops of the mountains, on the one hand, and the sea shores, on the other:—While there is a sea shore and a higher ground, there is that which is required for the system of the world: Take these away, and the world would remain an aqueous globe, in which the world would perish."

Yet it is obvious enough that before climate can work on rock surfaces, some of the latter must be raised above sealevel; once exposed they will probably consist of differing geological compositions which will withstand the attacks of weathering and erosion in different ways and at different rates. Most landforms must be considered to represent the interactions of their structure, stability relative to base level, the processes at work on them past and present, and the length of time for which those processes persisted. The complex nature of each of these parameters and the multitude of possible combinations of their variables ensure as regards surface shapes—or slopes, or whatever is meant by morphometry—that all forms of hillslope occur in all climatic environments.

The scientific analysis of the shape and spatial distribution of landforms is an enormous task still largely uncompleted yet it forms the basis of early climatic geomorphology. The later or latest trend is to determine

the exact cause and rate of the climatic process, and in turn to determine the relative importance of each major component of climate. At the same time the relative importance of the passive parameter, the rock structure, may also be revealed.

It is in this spirit that *Geomorphology and Climate* presents an analysis of the interactions between climatic processes and the shape of landforms. Written by fifteen well-known authors from Australia, Europe and North America, it can safely be said to be authoritative and certainly is neither dull nor dogmatic. It opens with a broad survey of the problems on the assumption that the central concern of climatic geomorphology lies in the extent to which variations in the elements of climate are reflected in geomorphic processes to produce distinctive suites of landforms. After a discussion of the traditional approaches by way of broad zonal patterns based largely on climate, vegetation and soils, the evidence of surface shape is shown to be inadequate as the sole basis for testing the influence of climate on landforms.

The following six chapters proceed in a logical sequence to discuss climatic interactions involved in rock weathering, mass wasting and the development of soil catenas and slopes. As would be expected of these experienced campaigners, the approach is modern, thorough and objective. For example, we learn that attempts to classify mass-movement types and frequency and their role in landscape development in terms of climatic parameters are doomed to failure; that, except in extreme situations, it is premature to relate climatic-geomorphological systems to soils; and that at medium or regional scales, slope forms cannot usefully be viewed as responses to climatic variables alone.

The next four chapters deal with water in hydrological slope models, with fluvial erosion rates, drainage networks, and, using West Malaysia as a detailed case study, with the role of climate in denudation. In these fields the influence of climate seems to be more securely established. For example, although several of the interactions are poorly understood and much more practical measurement is needed, the present findings and models of hydrological slopes are 'reasonably acceptable' and, moreover, within morpho-climatic zones the values of drainage density do reflect the climate.

Inevitably the discussion then turns, in chapters 12–14, toward lithology, which provides the framework and constraints in which climatic influences bring about the nature and rate of weathering, denudation and erosion.

There seems to be no doubt that geological factors may become dominant on a small scale and tectonic factors on a somewhat wider scale. The relative importance of rock composition is analysed with particular reference to granites, limestone, sandstones, schists and basalt. Just as the use of standard climatic means is being replaced by that of more pertinent extremes, so the use of crude rock types is being refined by the subtleties of microscopic analysis of rock composition and texture.

The concluding chapter gives a nicely rounded account of climatic factors in cirque formation. Not surprisingly, as cirques are often partly inherited from a pre-existing fluvial system and have close relationships to faults, the authors found it difficult to make generalisations on cirque variations in relation to climate.

This excursion to the snowline inadvertently draws the reader's attention to the absence of chapters on glacial, periglacial, and arid geomorphology. But the editor has already explained this omission. In his opinion, large-scale global, zonal or regional groupings, known as climatic geomorphology, reached their acme in 1950–65 and probably cannot profitably be taken much further until their broad assumptions have been fully tested by establishing the precise relationships between landform shapes and climatic parameters. Consequently this text is on geomorphology and climate.

Bear in mind, however, that in landform evolution the present dynamic processes are often not the key to the past, and that, as an American wit said, the landscape has been out of doors a long time. Because of wide swings in climatic zones during the Quaternary and the remarkable survival and reappearance of relic features from the more distant geological past, over large areas the present landforms are partly a product of past processes. But present processes do, however, provide the key to present denudation and therein lies much of the value of this collection of essays. An understanding of geomorphic processes and of their influence on the existing landscape is essential to Earth Sciences in an age when man's disturbance of the natural environment has become excessive. Consequently the advances in knowledge and experimental techniques discussed here are to be welcomed, especially as the clear exposition and frequent diagrams make them readily intelligible. □

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Climatic change for better or worse

Hans Mörtz

The Genesis Strategy: Climate and Global Survival. By Stephen H. Schneider. Pp xxi+419. (Plenum: New York and London, 1976.) \$14.95. *Forecasts, Famines and Freezes: Climate and Man's Future.* By John Gribbin. Pp. 132. (Wildwood House: London, 1976.) £4.95.

AUTHORED by one of the world's leading atmospheric research scientists who has specialised in the study of climate and its socioeconomic implications, Stephen Schneider's *The Genesis Strategy* has all the ingredients of a classic in its field. Nine chapters, ranging from strictly scientific aspects of atmospheric behaviour to the broad sociological spectrum of governmental strategy in the face of possible disaster, provide an immense wealth of food for thought. Written splendidly with the assistance of Lynn E. Mesriow, this book is likely to appeal most to those intellectuals who, despite having worked for years with logical reasoning and cold numbers, have maintained an open mind and heart for the real problems of this world. For the scientist, there is a wealth of useful references to literature on recent research in climatology and other related disciplines. Thirty-eight well selected diagrams provide the illustrations for key issues discussed in the text.

I will concentrate on the scientific atmospheric aspects of Schneider's work. A short account of recent climatic fluctuations leads up to the crucial question whether human activities have contributed to their occurrence. Inadvertent climate modification by man's activities is unquestionably gaining importance while present trends of the world's industrial activities continue. But, have we reached the point where large-scale climatic features can be produced? Although there is a general appreciation among climatologists of the basic problem (and it is a real problem), Schneider's emphasis and timing are likely to be regarded as overdone and premature. Nevertheless, in the spirit of the message conveyed by this book, there is justification for over-anxiety, as some consequences of inadvertent climate modification are irreversible once the point of no return has been passed.

One chapter is devoted to the problem "Climatic History—What if it Repeats?". Excerpts from well-documented accounts of historic climatic events provide a convenient introduction to this topic. The prehistoric 'palaeo'-climatic period, especially the occurrence of ice ages, is also briefly discussed. The lessons to be learned are manifold and important. Essentially, the study of past climates requires an interdisciplinary approach, including archaeology, geology, glaciology, history and palynology, among others.

A review of the theory of climate is presented. Here, the meteorologist reader will feel more on home ground, relatively speaking. Schneider places much emphasis on local changes of albedo on land surfaces due to differential land use, especially grazing of domestic animals. These albedo changes can be detected on photographs taken from Earth Resources Technology Satellites (ERTS). It is my opinion, however, that these are no more likely or unlikely to affect the large-scale circulation of the atmosphere than the condensation cloud trails produced by vast numbers of aircraft. Schneider does not consider the effects of the latter, whereas the bedouins are mentioned in three different contexts as possible culprits behind the Sahel drought disaster. Later in this chapter, it is conceded that climatic cause-and-effect linkages are very difficult to prove because there is still no adequate quantitative theory to explain any of the possible causes responsible for a particular climatic event.

Sixty pages of this volume deal with specific aspects of weather and climate modification. This is probably the most comprehensive presentation of topical aspects in this field available at present. In a world scarred with the effect of man's disastrous attempts to interfere with nature it is most satisfying to follow Schneider's thoughts on the advisability and wisdom of climatic control: "Control the Climatic Controllers, Not the Climate".

The climatic influence on food production, the present state and future needs of food supply for the world's population, and the dilemma which is likely to arise in the near future are portrayed with clarity and conviction in the focal chapters of this book. Following the pattern of climatic extremes which adversely affected crop production of the world's grain belts in recent years, the question of "can something be done?" looms between the lines.

Assuming that Stephen Schneider regards himself basically as an atmospheric scientist, the fundamental question arises whether scientists should pursue their professional activities

regardless or whether they should make an effort to ensure that the application of scientific knowledge to national and international problems is scientifically sound and morally justifiable. In writing this book, Schneider has emphatically affirmed his willingness to share the responsibilities towards the world. No doubt, the outspoken manner of his treatise will raise a number of eyebrows and tread on a number of toes. Perhaps, this is just what is needed in some circles of the profession which have become complacent over solving 'their' problems by finite difference steps—*ad infinitum*.

THE contents of the second book, *Climate, Famine and Freezes*, deal with two aspects: evidence for the notion that climate is changing, and scientific work dealing with this problem.

An outline of the large implications for today's economies of relatively small-scale climatic trends and fluctuations supports the view that climatic research is essential and a matter of urgency. Attention is drawn to the potentially high benefit-cost ratio for research in this field.

An account is given of recent temperature and rainfall trends, some of which have already infringed drastically on human life—for example, Sahel droughts, and Mediterranean region summer rains. Some of the statements and conclusions are fleeting and lacking in precision. The essential features of recent climatic trends, however, are well enough portrayed, and the recent drought (1975–76) in north-western Europe and the British Isles fits well into the picture.

On previous climatic changes, the author describes methods and techniques of obtaining prehistoric weather information indirectly (by proxy) from tree rings, sediment varves and pollen spectrum analysis. Attention is drawn to recent work in Africa linking equatorial rainfall with the extent of glaciation.

Implications of recent variations in the area covered by ice and snow in the Northern Hemisphere on the weather and climate, are discussed. Any hints at the likely causes of these changes must at this stage be regarded as pure speculation.

Gribbin devotes a chapter to the question of solar influences on weather—a most controversial and debated aspect of atmospheric science. Whatever the reader's view on this matter he must concede that the information given makes interesting reading.

Climatic trends of a long-term nature and their derivation from historical and prehistoric evidence are discussed. This work is gaining importance with regard to climatic forecasting as it delineates the boundaries within which the climate varied in the past. As one climatologist

put it: "What happened in the past can happen again". Unfortunately this theme leads Gribbin to speculate on solar-terrestrial weather relationships of a kind for which no scientific proof will be possible in the foreseeable future.

One of the most recently explored geophysical aspects of weather and climate is the Earth's magnetic field. Although this subject may be anathema to those meteorologists who believe that one can well manage without it, there is certainly justification for research, especially as good long-term records exist. At this stage of affairs, one simply cannot afford to reject outright a possible influence on, or association with, climate and its variations.

A better understood, but equally difficult, field of meteorology is the penetration of solar radiation through the atmosphere; or, looking at the other side of the coin, the loss of surface heating through interception of solar radiation by dust, clouds and aerosol in the atmosphere. The author correctly stresses the sensitivity of polar and sub-polar ice and snow cover to the delicate heat balance in high latitudes.

The question whether man's activities are changing the climate is dealt with in its widest terms. The author's treatment reflects the present majority opinion among scientists working in this field: although evidence for small-scale effects already exists (urban heat islands, rainfall modification, and so on), the large-scale atmospheric circulation is still overwhelmingly governed by "natural" events.

An objective and balanced view is taken in the discussion of the prospect for an early and sudden reversal to ice-age conditions. Major theories of global cooling are presented and put into perspective. One passage in the description of the astronomical theory of ice ages—"The most extreme effect by this process reduces the intensity of sunshine reaching the Earth by 30%"—is likely to produce an erroneous impression in the reader. Incidentally, Gribbin is in very good company when tackling this explanation, as Sir John Herschel may also be accused of misleading the readers of his *Outlines of Astronomy* (1875 edition) when he wrote: "... by concentrating half the annual supply into a summer of very short duration and the other half over a long and dreary winter, . . .". When Gribbin writes that the world's climate involves more complex factors than a simple dependence on variations in solar energy produced by changes in the Earth's orbital motion, he really puts the finger on the right spot.

Another important point is brought out in the discussion of the limitations to conventional forecasting techniques. Numerical computerised forecasting

techniques of the kind applied at present to the production of short-term (up to 72 h) forecasts cannot be stretched to cover longer periods while still providing acceptable results. Development of long-term forecasting requires the inputs of different kinds and timescales, including slow-acting feedback mechanisms, such as may be produced by changes in ocean surface temperatures. Present attempts at climatic forecasting are very much in the experimental stage.

Evidence for the association of grain and food production fluctuations with the sunspot cycle is scanty and covers too short a period. Surely a good number of the ups and downs of agriculture around the world are bound to coincide with sunspot maxima and minima. What about the remainder? More relevant and indeed important is the problem of crop failures due to weather extremes in our times when the world's

food reserves are nearly exhausted. Gribbin draws a far-reaching conclusion: that the economic policies of many countries may have to be altered.

In a postscript, the ultra-long aspect of changes in the Sun's radiation due to the Solar System passing through dust lanes in the spiral galaxy are considered. Gribbin's flare for an astronomical view on the problem is appreciated.

This book is a well written popular account of the state of the art of explaining the past and speculating on future climatic changes. It contains the essentials of present knowledge of this subject. Good bed-time reading for the layman and unbiased meteorologist. □

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Mathematical models in ecology

John H. Lawton

Theoretical Ecology: Principles and Applications. Edited by R. M. May. Pp. viii+317. (Blackwell Scientific: Oxford, London, Edinburgh and Melbourne, 1976.) Cloth £8.50; paper £4.80.

I ONCE heard Charles Elton say that the trouble with ecology was that it was "fun to do, but not very interesting to read about". This book makes it abundantly clear that ecology is now not only exciting to do, but also makes excellent reading.

The aims of the book are very simple: it seeks to review and draw together a whole series of mathematical models, to show how they can shed light on empirical observations and to examine some of the practical implications. In so doing, it occupies a niche between the numerous large general textbooks of ecology that have appeared recently, and the more technical literature that cascades on the world in the form of journals and specialist monographs. It embraces a range of subjects reflecting the core of current interest in theoretical ecology.

Certain relatively small areas receive scant or no mention (for example, optimal foraging theory, or spatial patterning in vegetation), and the book draws almost entirely on theoretical models that can be handled analytically. Large scale computer simulations

('dirty great' as opposed to 'little dirty') models figure hardly at all. It is also a theoretical edifice built on an inverted pyramid of biomass, with animal examples well in excess of plant examples. These are not criticisms.

The organisation of what the book does cover is conventional, and sensible. It starts with an examination of the dynamics of single species populations by May, and is followed by a review of the evolution of major population characteristics by Southwood. Between them, they illustrate particularly well something of the how and why a population's characteristics are as they are (in the resources allocated to reproduction, longevity, and so on), and how these characteristics influence its dynamics. Interactions between two populations are considered in the next four chapters, which deal first (by May) with some of the general features of predation, competition and, less conventionally, mutualism; and then in more detail with arthropod predator-prey interactions (by Hassell), plant-herbivore systems (by Caughley) and competition and niche theory (by Pianka). One of the more interesting things about ecology is the possibility of advance from a consideration of the dynamics of a single species or pairs of species and ignoring all the rest. It looks like cheating, but it also seems to work, as the interplay between theory and practice developed in these first six chapters clearly illustrates. In reality, perhaps many ecological systems are very loosely connected in terms of the major controlling links between species.

In certain cases, of course, it is misleading or nonsense to confine attention to single species, or pair-wise interactions. Various multi-species problems

are therefore considered in the next three chapters, which deal respectively with patterns in multi-species communities (by May), island biogeography and the design of nature reserves (by Diamond and May), and succession (by Horn).

The remaining four chapters would not normally be classified in the mainstream of theoretical ecology—friends or half-cousins perhaps, but not close family. In fact, they link rather well with the rest of the book, and serve to remind us that the traditional boundaries of the subject are in a state of flux. Wilson provides a brief overview of some of the central problems of sociobiology as he sees them and forces the reader to think about such problems as group selection, the evolution of sex, and adaptive demography. Gould expands the timescale of traditional ecological thought and attempts to bridge the ravine between palaeontology and ecology; and Cohen directs our thoughts inwards to the biological and ecological puzzles presented by our schistosome parasites. Finally Conway shows how ecological ideas can provide important insights into the behaviour of another group of nasty animals—insect pests.

Two criticisms are frequently levelled at multi-author texts: lack of cohesion and a large variance in performance. Neither can fairly be levelled at this book. Each author takes particular pains to draw on, and refer to other parts of the book. To give but two examples. Conway makes very successful use of the concept of the 'r-K' spectrum documented at length by Southwood, and Gould is able to use the species-area relationships that form the backbone of the chapter by Diamond and May, to provide a convincing explanation for one of palaeontology's biggest paradoxes—the Permian crisis of extinction. This interplay of ideas is obvious throughout the book. The papers do vary in quality, of course; all are worth reading, and most are of the highest standard. I found the paper by Caughley the most disappointing, in that it makes the least convincing contact of all between the real world and the models, but I suspect this is as much a fault of the lack of data as anything else.

This book will surely be essential reading for all serious students of the subject (both advanced undergraduates and postgraduates), and a valuable source of authoritative summaries and guide to the literature for teachers and research workers, for several years ahead. Not only will they learn a lot: they ought to enjoy it. I did. □

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Room with a view

K. G. Eldridge

The Natural History of Trees. (The World Naturalist Series.) By Herbert L. Edlin. Pp. xvi+269+66 plates. (Weidenfield and Nicolson: London, September 1976.) £10.

HERBERT EDLIN looks out at trees and forests of the world from the window of a country house not far from London. There is much to be seen and learned nearby, and much more to be remembered from his earlier experiences in other lands.

There are fine precedents for his approach to writing a natural history book: *The Origin of Species* was written from a house with a similar view. Edlin's latest book, however, does not present any grand concept or develop a thesis. It is descriptive, aiming to present factual information which might form the basis of ideas and rational action.

Administrators, politicians, some scientists and engineers, students, and other people who argue about the use of forest land, whether to chop trees down or grow more, often talk in ignorance of the basic biology of trees. This book will be a useful introduction to informed discussion.

Edlin deplores policies of single use of forests and advocates prudent multiple use. He does not like the current single-use ideas of wilderness, conservation, restricting access and allowing little or no management. Nevertheless he agrees that forest conservationists have reason to be active and angry. He shares their outrage at the single-use policy called "cut out and get out" which has exhausted much of the conifer forests of the west coast of North America.

People do not put a high value on their forests while wood and other products are available in unlimited amounts at no cost, like fresh air. The value of these commodities only becomes apparent as they are depleted or obliterated. With forests valued more highly and with a greater understanding of forest biology Edlin expects some of the dilemmas of the use of forest land to be resolved.

As a naturalist he has an obvious pleasure in trees and in understanding the natural processes of their life. His descriptive writing is attractive and clear, touching on every aspect of the

biology of trees both as individuals and in communities. Following the familiar pattern of the older textbooks of forest botany he describes the anatomy and development of seeds and seedlings, wood and trees, the ascent of sap, and the annual cycles of leaves and flowers. The descriptions are brief and accurate but the illustrations are inadequate. Good illustrations are essential to supplement the description of the cambium and how it makes wood, or of the guard cells of stomata and the way they let water vapour and air in and out of leaves.

Throughout the book Edlin shows a preference for writing of "purpose" rather than of "function", of "winged seeds designed for wind dispersal", as though the tree had decided and acted on what was best for itself. Such a teleological explanation seems simple, as it avoids the issue of evolution. Unfortunately it denies the reader the great satisfaction, which Julian Huxley and other splendid English writers on natural history gave us, of recognising adaptations as being the result of natural selection.

The descriptions of trees in communities in relation to soil, light and climate show how one kind of tree replaces another and the dependence of trees on their environment. The crowns of large trees in a forest are exposed to the macroclimate of the district, while the floor of the forest has a calmer, darker, more humid environment with less fluctuation in temperature. The author shows a special interest in the effects of wind on trees.

His outlines on forest soils and soil erosion give little hint of the vast amount of information on those subjects. In fact he could have mentioned several textbooks for every page of the whole book, but there are no references to any other publications.

With the scientific forester's approach of "wise management for sustained yield" he describes the many sorts of forests throughout the world and the ways they are used. In his account of the characteristics of broad-leaved and conifer forests of the Northern Hemisphere his explanations of the distribution of species are too brief to be satisfactory. One might agree at first that conifers predominate towards the Arctic because it is an advantage for a tree to be evergreen and ready to grow immediately the temperature is warmer than 6 °C. Edlin does not, however, discuss the rapid growth of the deciduous birch which is always present with the pine and spruce at the far northern timberline.

There is a similar lack of space to do justice to the complex ecology of the open savanna forests of tropical countries. Small trees grow in a grass-

land sustained by man-made fires, but when eucalypts are planted they soon outstrip the native trees. A recent explanation, which the author does not mention, is that the roots of foreign trees can penetrate a hard layer deep in the soil, which is impervious to the roots of native trees, and obtain water from greater depths in the dry season.

The material in the book is more about the trees and forests of southern England than elsewhere. Consequently its greatest appeal is likely to be to those readers close to the writer's familiar home ground who also would like a glance at living forests of the rest of the world.

Some of the most attractive parts of the book are those in which the writer gives an account from his personal experience of a forester's daily life in England, of a Malaysian rubber plantation and of rainforests. Edlin gives a lively description of the complex, diverse and changing tropical rainforest and notes with regret that the modern rainforest "is more likely to be raped than virgin".

Being an experienced writer in his field of popular forest science, Edlin

is concerned to be correct in his factual information. The few minor inaccuracies I noticed probably resulted from the brevity needed to fit such a large number of topics into 269 pages. For example, it is not correct to say the natural range of *Eucalyptus saligna* is in a Mediterranean climate. It is in an area of predominantly summer rainfall grading into a winter rainfall, Mediterranean climate at the southern end. Also, it is not correct to suggest that sulphite pulps are the main raw material for small paper bags, although they are sometimes used.

It was a curious decision in an otherwise well-produced reference book to have no suggestions for further reading to encourage the interest aroused in students. There should have been room for a bibliography and also a glossary. Nevertheless Edlin's book is readable and informative and will be a useful general reference for newcomers to the study of trees. □

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Broad canvas for cytoplasmic genetics

R. A. E. Tilney-Bassett

Cytoplasmic Genetics and Evolution. By Paul Grun. Pp. xi+435. (Columbia University: New York, July 1976.) \$31.25.

THE publication of a book in the field of extranuclear inheritance is a rare and welcome event. Paul Grun begins by establishing the physical properties of mitochondria and plastids, and their potential for genetic variation through mutation, segregation and recombination. A few of the more important mitochondrial and plastid enzymes are each briefly described, as if the author wished to emphasise that he had not completely neglected the biosynthetic capabilities. But, in my view, this was insufficient to serve much purpose and a greater attention towards clarifying the often quite complicated genetical accounts would have been more valuable. Nevertheless, I was impressed by the way in which the reader is briefly and carefully guided through the experimental procedures, and by the way the evidence is sifted and both sides of controversial issues discussed.

A large section on symbiotes and inherited diseases extends cytoplasmic inheritance into consideration of the relationships between bacteria and their insect hosts, spirochetes in *Drosophila*, killer parametia and killer yeasts, and the problem of cancers in vertebrate animals. Non-cancerous diseases are also discussed using the example of scrapie in sheep and Leber's optic neuritis in man to illustrate the importance of understanding the interactions between nuclear genes and transmissible factors. Yet, surprisingly, Grun fails to drive home the message that probably several other diseases might be better understood if regarded in this light. Having opened the door to discuss symbiotes, I was disappointed to find no mention of those remarkable chloroplasts that can exist endosymbiotically in the cells of some molluscs and other invertebrates.

The final of the four parts catalogues many examples illustrating the importance of the plasmon as the causative factor in determining some of the stranger facts of cytoplasmic genetics. Particular attention is given to the interaction between nuclear genes and plasmon factors, as for example in cytoplasmic male sterility, and to the evidence for genetic variability and evolutionary change within the plasmon. The chapters of this part add up to an extensive and thorough review reflecting the author's close familiarity with this little understood subject—a genetic backwater that needs this kind of publicity.

To bring together into one book such a wide range of genetic phenomena is a Herculean feat—over one thousand references are quoted. But Paul Grun is not content with pure description; he has attempted a modern synthesis in which he relates cytoplasmic phenomena to varying states of an overall evolutionary process. Whether we look at a parasite starting to invade a new host, or a long established organelle like a mitochondrion, there is a continuing process of genetic and physiological adjustment by which host and invader work out a strategy to their mutual advantage. Gradually, the successful parasite may become the universal symbiont and essential organelle. Initially the host may not be able to resist invasion, yet slowly and inexorably, through mutation and selection, the host nucleus gains control. At this stage any major disturbance to the harmonious relationship between the two is strongly selected against, and so the interaction between nucleus and cytoplasm becomes a restrictive force on evolutionary divergence. So close and so delicate is the interaction, that we find many examples of geographical races or neighbouring species, in both plants and animals, that fail to form healthy fertile hybrids because the hybrid nucleus is incompatible with the maternally inherited cytoplasm. In short, the cytoplasm has become an important force in evolution.

The evolution of a mechanism to limit the transmission of cytoplasmic factors to the female line is seen as a strategy by which the host effectively stops the recombination of cytoplasmic genes and thus gains mastery over their behaviour. Once sufficient control is acquired, the host nucleus may even take over some organelle genes as seems to have happened in the splitting of the cistrons for certain plastid enzymes between nuclear and plastid DNA, and in the reduction in coding capacity of vertebrate mitochondria compared with that of yeast or higher plants.

Although some areas are covered too thinly, the author's aim to paint a broad canvas covering the more important aspects of cytoplasmic genetics is generally successful. Above all, the illustrated book is written in an easy style, and it makes such fascinating and enjoyable reading that it should have a wide appeal to degree students in the biological sciences as well as to research workers. It is to be hoped that the high cost does not prove too great a discouragement for a book that deserves to be widely read. □

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Modern microbiology

Sydney Selwyn

The Development of Microbiology. By Patrick Collard. Pp. 212+xx photos. (Cambridge University: Cambridge, London and New York, November 1976.) £6.50.

THERE is probably no field of science or medicine which has a more fascinating and exciting history than microbiology. Paul de Kruif's classic, if over-romanticised, epic *The Microbe Hunters* has undoubtedly stimulated many young people to enter the medical and related professions during the 50 years since it was first published. The same source provided initial inspiration for the recent British television (BBC2) series *Microbes and Men*. The enthusiastic reception which greeted these dramatised sequences showed that the history of medical microbiology maintains a very wide appeal.

Paradoxically, however, this enthralling subject has received relatively little serious historical study. William Bulloch's *History of Bacteriology* stands alone as a detailed but idiosyncratic work. First published in 1938 it is now, sadly, unavailable. A briefer account by W. W. Ford, written in 1939, was followed more recently by W. D. Foster's general history. This contains a good survey of important French and German achievements of the nineteenth century but it neglects the earlier (and most recent) periods, and much of the less familiar British work. A very welcome event, therefore, is the publication of a new historical survey by Professor Collard of the Manchester Medical School. It arose from lectures to postgraduate students of medical microbiology and is admirably suited for such an audience. The cognoscenti, too, will find many delights but these will inevitably be tempered by disappointments especially in relation to the 'archaeological' period before Pasteur.

The book is divided in a very practical way into thirteen main chapters which consider in turn the development of such aspects as microscopy, microbial cultivation, chemotherapy, genetics and serology. The author has wisely resisted the temptation to deal chronologically with the entire subject *en masse*. At the outset he delineates four eras through which microbiology has passed: speculation, from 5000 BC

to 1675; observation, from 1675 to the mid-nineteenth century; cultivation, to the beginning of the twentieth century; and the era of physiological study, persisting to the present day.

Although this view has the merit of simplicity, it presents a static picture of a dynamic subject. In particular, it ignores the fact that the volume and quantity of speculative writing actually continued to rise steadily after 1675. The later landmarks include Benjamin Marten's book on consumption of 1720 and Richard Bradley's treatise on the plague of the following year, but hundreds of others could be cited, culminating, during the mid-nineteenth century, in the writings of Jacob Henle on infection in general, William Budd on typhoid and John Snow on cholera.

Perhaps surprisingly, the greater part of this work was British—as was a remarkable amount of more practical investigation. Microscopy begins logically with the elegant treatises on lenses by Roger Bacon and other Oxford philosophers of the thirteenth century, proceeding through the work of such forgotten figures as Leonard Digges, the elder, in the mid-sixteenth century.

The first practical microbiologist, however, was probably Thomas Moffett. This English physician, who is likely to have been the father of the celebrated "little Miss Muffet", not only described the infective cycle of silkworm disease, but also in 1589 provided a practical basis for a germ theory of disease with his study of the biology, pathology and treatment of scabies. Although Moffett is the undoubted founder of parasitology—and conceptually, of medical microbiology—he is totally neglected by the historians of these subjects. So also is one of the greatest theoreticians of all time, Francis Bacon, whose writings contain the roots of all that was to follow in microbiology. His assertion that an understanding of the processes underlying fermentation and putrefaction would be the key to the mysteries of disease and other "operations of nature" forms the basis for the later researches of Boyle, Willis, Pringle and their successors. Similarly, his perceptive remarks on the potential value of the new microscope lead directly to the work of that remarkable group of pioneer microscopists of the mid-seventeenth century—Mundy, Charleton, Power, Grew and Hooke. Only the latter has received any attention by historians of microbiology, and Professor Collard gives him merely passing mention.

Van Leeuwenhoek receives a more adequate amount of space, but some misconceptions need to be dispelled. He did not arise *deus ex machina* but

his single-lens microscopes are perfect replicas of the type fully described ten years earlier by Robert Hooke, who also provided the idea of modified dark-ground illumination which probably determined van Leeuwenhoek's later successes. The Dutch draper was, moreover, incapable of producing the drawings contained in his famous letters to the Royal Society. Clifford Dobell, his reverential biographer, describes the mediocre quality of the work done for van Leeuwenhoek by his draughtsmen. So much so, that Professor Collard identifies "streptococci" from a sketch intended to portray, with a series of dots, the track of a mobile bacillus.

There are few such problems of interpretation in the modern period, although the story of Fleming's rediscovery of penicillin needs overhauling following Ronald Hare's revelations. Also Domagk's prontosil red was virtually inactive *in vitro* and was relatively ineffective against pneumococci *in vivo*. These and other minor errors should not be allowed to dissipate our genuine gratitude to the author. He has written the best available account of the development of modern microbiology. □

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Imaginative processes come of age

P. W. Hawkes

Optical Information Processing. Edited by Yu. E. Nesterikhin, G. W. Stroke and W. E. Kock. Pp. xiv+401. (Plenum: New York and London, 1976.) \$39. *Digital Picture Processing.* By A. Rosenfeld and A. C. Kak. Pp. xii+457. (Academic: New York and London, 1976.) \$28.50. *Picture Processing and Digital Filtering.* Edited by T. S. Huang. Pp. xiii+289. (Springer: Berlin and New York, 1975.) DM79.80; \$34.40. *Principles of Optics.* By M. Born and E. Wolf. Pp. xxviii+808. (Pergamon: Oxford and New York, 1975.) £10.50; \$22.50.

TEN years ago, digital image processing hardly existed and optical processing was in its infancy. Microscopists, radio-graphers, archaeologists studying aerial photographs and a host of other users of images of some kind were well aware of the shortcomings of their interpretative techniques and they at best overcame or at worst disguised them by slowly accumulated experience. Nevertheless, optical processing cannot truly be described as a new subject, for it consists in combining a thorough knowledge of the optics of the image-forming process with some ingenious optical designing intended to avoid the most troublesome of the inherent limitations, probably at the expense of worsening less harmful ones: apodisation is an obvious example. The sudden interest in optical processing in the 1960s was stimulated by the arrival of the laser, which permitted essentially coherent processing, although complementary incoherent techniques have also emerged or been reconsidered in the general revival of studies in this domain.

Digital image processing is, on the other hand, a genuinely new subject, which could not exist before fast computers with very large memories became widely disseminated. To a computer, an image is no more than a two-dimensional array, usually as large as possible, of real or complex quantities; and the challenge of computer processing is a complex one: the discrete array must represent the recorded image, however degraded this may have been in the image-forming process, as faithfully as possible. This means studying the properties of the photographic medium and the optical properties of

the microdensitometer (unless the image is transmitted sequentially, as in a scanning device for example). Optimum strategies must be devised for rendering the discrete image better, in some carefully defined sense, or more informative, in the sense that the eye can recognise detail of interest more easily or that a quantitative measure is obtained of some property about which only qualitative information had been available. Finally, the processed image must be made visible preferably in a way that makes obvious its superiority over the original.

A number of books and review articles on coherent optical processing, many devoted mainly to holography, have appeared over the past few years and the volume by Nesterikhin *et al.*, which contains the seventeen papers delivered at a US-USSR joint seminar in 1975, is an uneven addition to this body of literature. In particular, some authors have been at pains to situate their work relative to earlier contributions, whereas others merely give a bald account of their own research; since there seems to have been no length limit—one chapter, an excellent one on the information content of diffraction patterns by B. J. Thompson, is 34 typed pages long—this approach is difficult to forgive. There are chapters on both the technology and the theory of optical processing, ranging from one on materials and devices by Casasent and another on holographic memories by Vander Lugt to a survey of speckle by Goodman and a discussion of holographic memory devices for specific integral transforms by Gibin and Tverdokhle. It is useful to see the lines which Russian scientists are following, as the only other sources of Eastern material, original articles apart, are the occasional *Sborniki* that have been compiled. Finally, a word about the editing: the book has been offset directly from typewritten texts submitted by the authors. For the price, the editors could surely have had the text retyped uniformly which would not only have produced a less scrappy-looking text but also have permitted genuine editing: the removal of such bizarre transliterations as “the Fuco-Hilbert transform” for example and the provision of some assistance to authors whose English was shaky.

In the field of digital processing, a torrent of books, many of a very high standard, has recently flooded on to the market, supplementing and replacing the excellent early texts by Andrews and Rosenfeld (by ‘early’ in this context we mean 1970 and 1969, respectively): indeed, the volume by Rosenfeld and Kak supercedes Rosenfeld’s earlier book, and a new and expanded edition of Andrews’ text is promised for the new year. The volume

by Rosenfeld and Kak and that edited by Huang contain essentially similar material, concerned with the mathematical representation of images in discrete form, compression of the information, often highly redundant, contained in them, the enhancement of contrast and the restoration of degraded pictures and some pattern recognition techniques; the latter are, however, much more fully described in other recent books not noticed here.

Picture Processing and Digital Filtering contains six contributions: a clear survey by Huang, putting the detailed chapters in perspective; an extremely good account by Andrews of most of the two-dimensional transforms in algebraic terms; separate chapters on recursive and non-recursive filters by Read, Shanks and Treitel, and by Fiasconaro, respectively; by far the best available account of mathematical techniques for image enhancement and restoration by Frieden; and a good account of the effect of noise, firmly related to the type of equipment used at present in image processing, by Billingsley. All these chapters are extremely clearly written and above all, given the novel status of the subject, they are written so that the techniques can be used and developed for a wide range of possible applications. The only disappointing contribution is that on recursive filtering, as yet a rather shadowy aspect of image processing, in which the authors never really explain what their techniques are for. But we should probably be grateful for any extended essay on this topic, whatever its shortcomings. The chapters by Andrews and Frieden are masterly and would alone make the book highly desirable.

The book by Rosenfeld and Kak is also very good indeed; although a direct competitor, in the sense that it has a great deal of material in common with Huang’s volume, the tone and approach are subtly different—due no doubt to Rosenfeld’s preoccupation with pattern recognition and picture descriptors during the past few years. The homogeneity of the writing is pleasant after the inevitable unevenness of the multi-author volume, and Rosenfeld and Kak have devoted a great deal of space and effort to ensuring that the reader grasps the purpose of the many techniques described. Furthermore, whereas Huang’s book has the merit that a great many techniques are described by their originators, Rosenfeld’s has the complementary merit of comparing and contrasting the same techniques from another viewpoint. In conclusion, most workers in this field will want to have access at least to both of these volumes; newcomers are probably better served by Rosenfeld and Kak’s *Digital Picture Processing*.

Finally, we turn to *Principles of Optics*; it is always a pleasure to praise this book, now in its fifth edition. Most of the earlier 'editions' were scarcely more than reprints, as was made clear in the prefaces. But in the fifth edition, much of the material on the optical properties of metals has been rewritten, where the original text was misleading. Although sympathising entirely with Professor Wolf's reluctance to tamper with the text piecemeal in an attempt to bring it all up to date, it would be particularly pleasing if he consented to incorporate into the sixth edition the new material recently published by his own group in the field of partial coherence and the radiometric quantities. Even so, it remains a near-definitive account of classical optics at a very reasonable price. It is included in this group of books on image processing since a thorough grasp of the chapters on image formation at least is indispensable for anyone seriously embarking on digital or optical processing.

How far has image processing got

in practice? In some respects, the mathematics has outstripped the practical applications with the result that the same test images tend to recur repeatedly after undergoing a wide variety of treatments. Nevertheless, the volumes on digital methods may leave the misleading impression that theory and practice are at present far apart, for there is little mention of specific applications: X-ray tomography, for example, or three-dimensional reconstruction of electron micrographs. Despite this, the possibilities are very exciting: solution of the phase problem; extrapolation beyond the classical limit of resolution; the acceleration of picture processing using fast transforms other than the FFT; and many others. The second decade will undoubtedly witness as many innovations as the first, no less unpredictable. □

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Proprioception

J. W. S. Pringle

Structure and Function of Proprioceptors in the Invertebrates. Edited by P. J. Mill. Pp.xviii+686. (Chapman and Hall: London; Distributed in USA by Halsted Press; August 1976.) £19.50.

A BIOLOGIST considering whether to read (or purchase) a volume with multiple authorship does well to consider not only the field covered but also the standing of the contributors. The editor's tasks is like that of a building contractor: he cannot construct well without good materials. Of this book it can be said at the start that the authors are well chosen. They come from Australia, France, Germany and the USA as well as from the UK, and have all developed their insight into the subject by original contributions to it.

In 1938, I published what may have been the first paper on Proprioception in Arthropods. Now, 38 years later, 419 out of the volume's 686 pages deal exclusively with this Phylum, and as much is known about them as about vertebrate animals. The diversity of form and function is much greater. A definitive textbook on vertebrate proprioceptors has been written by one man; for invertebrates the task requires a contributory volume and an editor like this one, prepared to identify the most qualified contributors and to perform such integration as is possible.

The chosen definition of 'proprioceptors' is not Sherrington's—his task was easier because he was considering only the vertebrates; this volume covers all "sense organs capable of registering continuously deformation and stresses in the body due to the animal's own movement or to its weight or to other external mechanical forces". So defined, the vertebrate semicircular canals, otoliths and lateral line would be proprioceptors, which may seem absurd. But such a wide definition is indeed necessary for invertebrates, and if this volume is nearly a textbook of invertebrate mechanoreception it is no less valuable as that.

Laverack's Chapter 1 is largely a structural review and well illustrates the diversity. In Chapter 2, Fields, in a review of all aspects of the physiology of the crustacean muscle receptor organ, includes a clear statement of the evidence for a causal relationship between the electrogenic sodium pump and adaptation of impulse frequency, the latest demonstration of the value of this preparation for basic sensory physiology. Our knowledge of the mode of action and behavioural utilisation of this organ is at least as complete as for the vertebrate muscle spindle. Bush follows this with an account of another preparation important to general physiology—the only sense organ known to operate without impulses.

In Chapter 4, Finlayson gives us a comprehensive review of the multiterminal neurone system of arthropods, the importance of which has only recently been fully appreciated. It seems to be responsible for the overall co-

ordination of movement necessary in a primitively soft-bodied multisegmental animal and to provide the main feedbacks by which the intrinsic motor patterns are adjusted to a variable environment. But neither in locomotion nor in the specialised mouthpart and gut receptors (chapter 5 by Wales) is it possible yet to decide whether these multiterminal type II receptors and the chordotonal type I receptors (chapter 6 by Mill) serve a distinct or a complementary function in proprioception.

At the base of the decapod leg are two peculiar groups of internal chordotonal organs which neither cross a limb joint nor are attached to a muscle or apodeme. In chapter 7, Clarac considers the probable role of these organs in the autotomy reflex. The arthropod section of the volume ends with a long chapter by Wright on limb and wing receptors in insects, chelicerates and myriapods, a detailed description by Moulins of the ultrastructure of chordotonal organs and a valuable chapter by Macmillan on apodeme tension receptors. The diagram which this last author gives comparing vertebrate and arthropod proprioceptive systems will no doubt be much used by lecturers, but ought to be viewed with caution in the present state of our knowledge.

As Dorsett points out at the beginning of chapter 11, proprioception in many soft-bodied invertebrates is not the well-differentiated system found in vertebrates and arthropods possessing a rigid skeleton. Most of the locomotor activity of these animals is stereotyped and is pre-programmed in the central nervous system. Orientation to gravity by means of statocysts is, however, widespread and is included under the definition used for the volume; but proprioceptors *sensu stricto* have only been established in a few cases. Spatial equilibrium in arthropods (chapter 12 by Sandeman) needs separate treatment since the necessary information may be obtained, without a special sense organ, by monitoring the relative position of the parts of the body under linear and angular accelerations. Molluscs (chapter 13 by Budelmann) usually have statocysts.

The final chapters of the volume deal with the processing of proprioceptive information. Wells considers its role in learning; Mill and Price give the necessary theory of feedbacks; and Wiersman writes generally on various relevant topics. The editor's final brief contribution picks up some loose ends and highlights residual problems.

Altogether a most useful book, well produced and not unduly expensive. □

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American response to Darwin

David L. Hull

Darwin in America: The Intellectual Response, 1865-1912. By Cynthia Eagle Russett. Pp. ix+228. (Freeman: San Francisco and Reading, May 1976.) Cloth £6.30; paper £3.20.

RUSSETT'S well-balanced account of the intellectual response to Darwinism in America further enhances the richly illustrated picture which we already have of the Darwinian revolution. As Russett would be the first to agree, her book could have been entitled just as readily *Spencer in America* because the authors which Russett treats were as likely to have their minds blown to bits by reading Spencer's *First Principles* (1862) as by Darwin's *Origin of Species* (1859). The genuinely scientific character of Darwin's theory legiti-

mised Spencer's Cosmic Philosophy, but the actual content of Darwin's theory had as little impact on its converts as on its opponents.

Russett deals exhaustively with all aspects of 'Darwinism' in America—scientific, philosophical, religious, social, historiographic, economic and literary. In spite of the opposition of the most powerful scientist in the United States, Louis Agassiz, and perversely to some extent because of it, evolution was almost universally accepted by American biologists within a decade after publication of the *Origin*, but the 'evolutionism' which became so popular tended to be Lamarckian and teleological—partly for scientific reasons, partly theological. Theologians were not alone in viewing the death of teleology as the death of God. Yet in slaying Paley, Darwin slew a corpse. Darwin merely shocked nineteenth-century intellectuals into realising how vacuous teleology had become. The Metaphysical Club at Cambridge was the focus of evolutionism among American philosophers. As might be expected, John Fiske, the most enthusiastic evolutionist, was a Spencerian. More serious philosophers such as William James, John Dewey, Josiah Royce, and Chauncey Wright, however, viewed Spencer as so much rubbish which had to be cleared from their path before proceeding on their way. Spencer was the "philosopher whom those who have no other philosopher appreciate" (p17).

The most interesting sections in Russett's book are those in which she deals with Social Darwinism and its effects on such novelists as Jack London, Frank Norris and Theodore Dreiser. Rugged individualism, socialism, aid to the poor, free will and determinism, nature-nurture, the relative superiority of the races, group selection and altruism—it is all here but presented more bluntly and at a safer distance than it is in the current brouhaha over sociobiology. Darwin also legitimised the 'historical method', which for Dewey was the discovery of the particular sequence of conditions which brought about a particular natural phenomenon, for Henry Adams the search for the one great law of history. Adams found his great law in a combination of thermodynamics and the law of inverse squares. Not only was the universe running down, it was doing so logarithmically. The religious phase of human history had lasted 90,000 yr, the mechanical phase would last 300 yr, the electrical 17.5 yr, and the ethereal phase but 4 yr.

Russett's prevailing concern is the interconnections between intellectual disciplines. For example, if Darwin and Wallace actually borrowed the notion

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of natural selection from Malthus's *Essay on Population* (1798), then the alacrity with which economists like Thorstein Veblen read the basic principles of evolutionary theory back into human affairs was understandable. A quick glance at Malthus's *Essay*, however, brings the argument full circle because in it Malthus justifies his views about human population by reference to biological species in a state of nature. As complex as the interconnections between intellectual disciplines may be, Russett believes that the content of scientific theories can colour the general intellectual climate and *vice versa*. Her final chapter concerns the blow which Maxwell's equations, quantum theory and relativity theory dealt to the absolute, certain, deterministic, mechanistic worldview initiated by Newton.

The book is illustrated with the usual portraits as well as with extremely well-chosen cartoons and paintings. Some of these illustrations convey messages much more powerfully than any printed word could hope to do; for example, the contrast between 'Nature red in tooth and claw' and the cloying romanticism of Albert Bierstadt's painting of the Rocky Mountains. My only criticism is that Russett completely ignores the really excellent scholarship which has been produced during the past dozen years or so on Darwin and Darwinism. If she has read any of the more recent work, she does not mention it. □

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Photo: Mansell Collection, London, UK

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review article

Products of the major histocompatibility complex and their relationship to the immune response

Alan Munro & Susan Bright*

The genes of the major histocompatibility complex were first known for the part they played in transplant rejection. Recently, however, it has become clear that the products of that region have an important part to play in the control of the immune response, through their effects both on cooperative and on aggressive interactions between cells. It is now possible to guess at the mechanisms which may underly the association of some major histocompatibility antigens with disease.

IN mammals there seems to be a single genetic region which has a major influence on graft rejection and is known as the major histocompatibility complex (MHC). In recent years there has been increasing interest in the products of this genetic region because of their importance not only in transplantation but in many other aspects of immunology¹⁻³.

The MHC is divided into different regions mainly on the basis of genetic recombinants⁴. The probable arrangement of these regions in mouse and man are shown in Fig. 1. In this review we want to discuss the properties of the MHC as a whole, in particular its unusual genetic properties and the association of certain alleles in the MHC with diseases in man. In order to do this it is essential first to discuss the properties of the genes which lie in the different regions of the MHC, in terms of their genetic properties and in terms of the molecular nature and suggested functions of their products. In this connection it is helpful to compare and contrast the properties of corresponding genes found in mouse and man. We shall then return to the problem of disease association with HLA in man and finally discuss the possible reasons for the unusual genetic properties of the region as a whole.

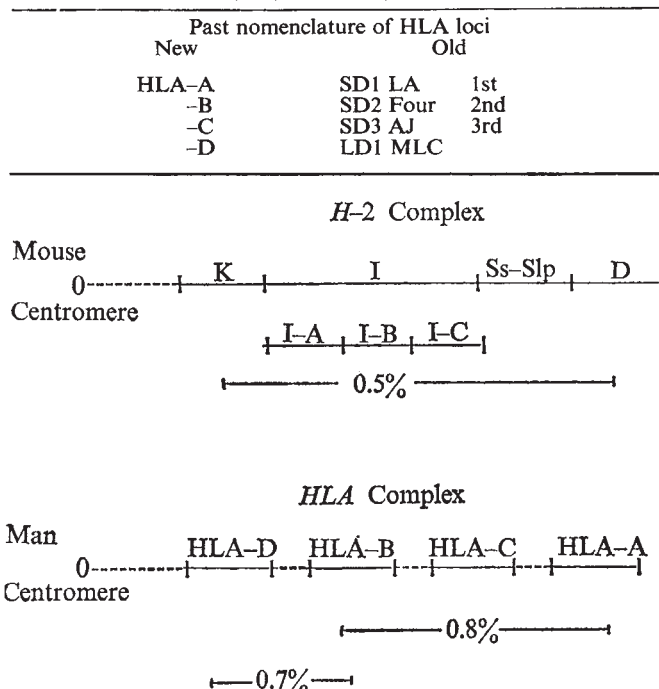
It is important to have some feel for the size of the MHC and hence a crude estimation of the number of possible genes involved. The MHC covers a length of DNA equivalent to 0.5 recombination units in the mouse¹ and greater than 1.0 in man^{5,6}. The low frequency of recombinants between the different loci in the MHC means that in most cases offspring inherit from each parent all the alleles found on one of the parental chromosomes. Such a set of alleles is known as a haplotype. It is not known how much of the DNA in the MHC codes for protein. The amount of DNA in a piece of chromosome of this size could, however, code for as many as 2×10^3 polypeptide chains⁷. It is important to remember that each region probably contains several genes. When more than one property is assigned to the products of a particular region then it does not necessarily follow that these properties belong to the same gene product. In time each region may be further divided as new recombinants are identified.

The classical histocompatibility antigens (CHAGS)

Historically the first products of the MHC to be characterised were those from the K and D regions in the mouse and the HLA-A and -B loci in man^{1,8}. The products from these regions have many biochemical and functional properties in common and seem to form a group of related molecules, which we shall call CHAGS.

The CHAGS can be detected by using antisera induced by the introduction of histoincompatible cells into recipients. In both species the antigens detected by such antisera are found on all tissues except possibly early embryonic tissue and mature human red blood cells. Individual cells

Fig. 1 Diagrammatic representation of the MHC of mouse (H-2) and man (HLA).



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from a heterozygous animal express the alleles from both chromosomes while individual sperm seem to carry the CHAGS and other MHC gene products coded for by a single chromosome^{9,10}.

Using different antisera people quickly realised that there were many alleles at each locus. In the mouse each allele of the K and D locus is described by a series of numbers representing individual determinants which fall into two categories; a private specificity which is generally unique to that allele and a set of public specificities which occur in more than one allele. Both types of determinants are on the same polypeptide chain¹¹⁻¹³, although there may be possible exceptions to this rule^{14,15}.

In humans the alleles are typed by using specially selected antisera which probably recognise the equivalent of only the private specificities in the mouse. Consequently each allele is described by one number, for example, HLA-A1, HLA-B5. There is increasing evidence for the existence of public specificities in man which may account for some of the anomalous patterns of reaction found with many anti-HLA antisera^{16,17}, for example 4a and 4b. It is not known if the present typing methods for HLA-A and HLA-B private specificities are adequate for transplant and disease association studies or whether typing for public specificities should be included. This is particularly important as private specificities in mouse and man are not always associated with the same public specificities respectively^{16,18,19}. It is worth noting that some human specificities of a public type are found in other species²⁰.

By using antisera to the CHAGS to monitor the purification, it has been possible to isolate membrane proteins carrying the corresponding antigenic specificities²¹. In both human and mouse these are glycoproteins of molecular weight ~ 45,000. The allelic forms differ in the amino acid sequence and it seems that the carbohydrates are not responsible for the allelic variants^{21,22}. Amino acid sequence data of the N-terminal ends²³⁻²⁶ have confirmed the structural homology of the K and D region CHAGS previously suggested by shared public specificities²⁷. There is also marked sequence homology between HLA-A and HLA-B N terminal fragments^{28,29}, although as yet the serological evidence for such a homology is weak^{30,31}.

There is structural homology not only between the CHAGS from one species but also between the N-terminal fragments of the CHAGS from different species. This homology lends weight to the suggestion that one is working with homologous systems; but the interspecies homology is less marked than that homology within species, and this raises interesting evolutionary questions. It would seem that speciation preceded duplication to form multiple loci. Ways in which this may have been achieved have been discussed at length elsewhere³².

The human and mouse CHAGS have other features in common. Both are found on membranes non-covalently associated with β_2 microglobulins³³⁻³⁶, remarkable proteins of molecular weight 12,000, which have 30% homology with an immunoglobulin domain^{37,38}. In both animals it seems that the carboxy-terminal end of the CHAGS is involved in attaching the molecules to membranes. A carboxy-terminal fragment of about molecular weight 8,000 is lost if proteolytic enzymes are used to solubilise the molecules from the membrane. The previously isolated polymeric forms of purified human CHAGS are now thought to be artefacts caused by the extraction procedures^{39,40}. Further it has been shown that the allelic products of each locus move independently on lymphocyte membranes^{41,42}. Consequently the molecules only interact with each other very weakly if at all on the surface of cells.

The independent movement of certain determinants on human lymphocytes supports the suggestion that there is a third locus in man, HLA-C, the products of which are

like HLA-A and HLA-B in their tissue distribution and association with β_2 -microglobulin^{33,43}. The relationship between HLA-C and the other two loci (HLA-A and HLA-B) is not yet known, particularly as HLA-C seems to be weakly immunogenic compared to HLA-A and B⁴⁴. The association of the CHAGS with β_2 -microglobulin has led to much premature speculation on the possible evolutionary relationship between CHAGS and immunoglobulin. The structural homology between β_2 -microglobulin and immunoglobulin is such that it is reasonable to assume that β_2 -microglobulin has the same overall three-dimensional structure as a variable region or constant region domain⁴⁵. Immunoglobulin domains readily interact to form dimers, and it has been suggested that the binding of β_2 -microglobulin to the CHAGS occurs by way of a similar mechanism. If this is so there should be a region with homology to an immunoglobulin domain in the sequence of the CHAGS. So far there is no evidence for this though the sequence data are still very limited.

Genetic properties of the CHAGS

We want now to discuss the exceptional genetic properties of the CHAG alleles, which set this system apart from most other known allelic systems. Within a population certain pairs of alleles, one from the A and one from the B locus, occur as haplotypes with a frequency which is greater than or less than that expected if the alleles were randomly associated in the population. This is known as linkage disequilibrium. For example, the haplotypes A1, B8 and A3, B7 are more common in the Caucasian population than would be expected from the frequencies of the individual alleles. Linkage disequilibrium can occur when there has been insufficient time for equilibrium to be established in the population concerned. It is generally thought that this is not the case for the CHAGS in most human populations⁴⁶. Alternatively, linkage disequilibrium can be caused by selective pressures operating in the population to hold certain pairs of alleles on the same chromosome. In different populations the HLA alleles occur with different frequencies and different haplotypes can be found in linkage disequilibrium in different races⁴⁷, which suggests that the selective pressure to establish linkage disequilibrium may be different in each race. Further these pressures need not operate on the CHAGS themselves but on genes on either side of them.

Another unusual feature is that most of the twenty or so alleles at the A and B loci have similar frequencies to each other in the Caucasian population. Selective pressures are required to account for this balanced and extensive polymorphism, for example through favouring heterozygosity^{7,48}. If selection operates through resistance to diseases then such pressures may no longer apply to populations treated by modern medical techniques. In Caucasian families now, segregation of HLA-A and HLA-B obeys the Hardy-Weinberg equilibrium^{49,50}, possibly reflecting the absence of selective pressures in western societies. It is interesting that in at least one population (Tuaregs) there is evidence for heterozygote advantage⁵¹. Because of linkage disequilibrium in the MHC the selective pressures to establish balanced polymorphism of the CHAGS may operate on linked genes, for example, the immune response genes (see later).

In this connection it has been suggested that heterozygosity of H-2 in wild mice may be promoted by the linked *T/t* complex⁵², which is said to control surface structures important in development⁵³. There are lethal recessive alleles (*t*) of several loci in this complex. Two unusual properties of these *t* alleles could lead to extensive heterozygosity at H-2. First, male mice carrying *t* alleles transmit the mutant form to their offspring with an exceptionally

high frequency. Second, many of the *t* mutants suppress recombination over a length of chromosome extending to the H-2 complex. This provides a mechanism by which H-2 heterozygotes are favoured as *t* mutants are lethal in the homozygous state. The *t* mutants are common in wild mice and can be found in as many as 40% of certain populations¹.

There is no evidence that a similar *T/t* complex exists in man; in fact there is evidence to the contrary. The properties of the *t* alleles are such that one would expect to see diversion from Mendelian segregation of the MHC. This has not been observed, at least in Caucasians. It thus seems likely that if the *T/t* complex does exist in the human then it does not bear the same relationship to the MHC as in the mouse. Alternatively the human *T/t* complex may not have the same unusual properties as the mouse complex.

Cytotoxic T lymphocytes and the CHAGS

Although in the past various biological functions have been suggested for the CHAGS^{7,18,52}, there has been little evidence to support any of them. Recent experiments have shown that the CHAGS are in some way involved in the interaction of cytotoxic T cells with their targets. In mouse and man reaction against a foreign MHC, for example in a mixed lymphocyte reaction, generates cytotoxic T cells⁵⁴⁻⁵⁸. In general the generation of such cytotoxic T cells requires cooperation between different populations of T cells: helper T cells which respond to differences in the I region (or HLA-D), and the cytotoxic T cells which respond to differences in the CHAGS^{59,60}. The cytotoxic T cells generated in a primary reaction of this type will kill target cells carrying the CHAGS to which they have been activated. In mice, for example, T cells activated to the H-2^s antigens will kill only target cells carrying an H-2^s CHAG at either the K or D region: the nature of the I or S regions is unimportant. Thus it seems that the CHAGS are the antigens with which such cytotoxic T cells interact. This restricted specificity of cytotoxic T cells must be a consequence of either a restricted range in specificities of the receptors for antigens on cytotoxic T cells (which seems unlikely) or a restriction imposed by the nature of the assay for cytotoxic function. In this context it is interesting that target cells lacking CHAGS can be killed by cytotoxic T cells only in the presence of concanavalin A (con A)⁶¹ a substance which may bypass the structures involved in normal recognition mechanisms.

It has been suggested that allogeneic responses are biologically important, for example, to prevent transmission of tumours between individuals⁴⁸. However, the successful survival of inbred strains of animals makes a biologically important role for such reactions unlikely and they probably occur only in artificial situations. Consequently the fact that the antigenic targets of cytotoxic T cells generated in an allogeneic reaction are the CHAGS is of little general significance. In mice, however, it has also been shown that in reactions of cytotoxic T cells to surface antigens other than CHAGS, the nature of the K and D region gene products on the target cells is still vitally important. Cytotoxic T cells specific for a variety of different antigens have been used in these experiments including, (1) viral antigens on virus infected cells⁶²⁻⁶⁹, (2) histocompatibility antigens other than the K and D region molecules^{70,71}, and (3) haptens on chemically modified cells⁷²⁻⁷⁸. In each case the cytotoxic T cells are lytic only if the target to be lysed carries the specific antigen and also the same K or D region molecules as the original stimulating cells. For example cytotoxic T cells from H-2^k mice infected with vaccinia virus will kill H-2^k fibroblasts if they are also infected with vaccinia, but not virus-infected H-2^b cells. But because of the partial homology between certain CHAG alleles (as reflected in their shared public specificities) it is not sur-

prising to find a certain amount of cross reactivity in some cases^{68,69}. (This is also seen with cytotoxic T cells generated by an allogeneic reaction^{75,76}.) It is not at all clear why both stimulating and target cells for cytotoxic T cells must express the same K or D region molecules. It is even possible that the K or D region molecules involved in some of these cytotoxic reactions are not the CHAGS but products of closely linked genes^{57,58,67,68,75}. Further, it is possible that the effects seen with different antigens, although similar in appearance, are different in origin. This may be particularly true for hapten modified cells.

Two explanations have been put forward to explain the above phenomenon. The first is based on the proposition that the specific antigens cause a modification of the CHAGS. According to this theory, which is known as the "altered self" hypothesis, the modified CHAGS are the structures that are recognised by the cytotoxic T cells. For example, virus proteins on the surface of cells might combine with the CHAGS and the complex become the new antigenic determinant. Alternatively the viral proteins might interact with the CHAGS, causing the CHAGS to alter their shape and hence become foreign antigens. It is hard to imagine in molecular terms how these alterations are brought about, particularly when the specific antigens concerned are the minor transplantation antigens rather than viral proteins. The fact that about 10% of unprimed T cells react to all the antigens in a single foreign MHC⁷⁹ has led to the suggestion that T cell receptors are designed to react with foreign MHC antigens. This would support the "altered self" hypothesis, particularly if the modified CHAGS were to resemble the CHAGS found in other members of the species⁸⁰. It is not known, however, if unprimed cytotoxic T cells show a similar high percentage of reactivity to a set of allogeneic CHAGS.

The second explanation proposes that cytotoxic T cells require dual recognition of target cells through (1) specific antigen and (2) CHAG determinants to kill the targets. This hypothesis takes various forms depending on the nature of the proposed recognition of CHAG determinants. It now seems unlikely that this is through a requirement that CHAGS on cytotoxic T cells should be identical to those on their target. Thus H-2^k cytotoxic T cells prepared from H-2^k-H-2^d chimaeric mice infected with lymphocytic choriomeningitis virus, killed with equal efficiency H-2^k and H-2^d virus-infected target cells. Supporters of this type of dual recognition hypothesis can still argue that the important requirement is that the target cells should carry at least some of the CHAGS which were present at a critical stage of the maturation of the cytotoxic cell. The cytotoxic cells in a chimaeric animal would mature in the environment of H-2^d and H-2^k cells and hence kill both types of target.

Current opinion favours a dual recognition hypothesis in which cytotoxic T cells recognise their targets by two independently coded receptors, one of which reacts with the specific antigen and the second with a CHAG determinant⁸¹. By this hypothesis the restriction of cytotoxic T cells recognition lies in the narrow range of structures (CHAG determinants) with which the hypothetical second receptor reacts. It is difficult to visualise a molecular mechanism by which a family of receptors reacts specifically with CHAG determinants and no other antigens.

A third form of the dual recognition model hypothesises that a cytotoxic T cell recognises its target through a single receptor which reacts with specific antigen and, through cross reaction, with a CHAG determinant on the target. Some receptors on a cytotoxic T cell would react with the target through specific antigen while other identical receptors on the same cell would react with CHAG determinants on the target. This hypothesis implies that such dual interaction is not only important for cytotoxic

function, but necessary for the proliferation of cytotoxic T cells so that only clones carrying the appropriate cross-reacting receptors are selected.

In summary, much is still to be learned about T-cell recognition and cytotoxicity and no wholly satisfactory explanation exists for the restriction seen in these systems. It is not yet known if such restriction applies to cytotoxic T cells in man.

Introduction to the I region

The I region of the mouse and the region around the HLA-D locus in man have much in common. Both were initially studied by using a mixed lymphocyte reaction (MLR) as a means of detecting antigenic differences. (The so-called lymphocyte defined or LD antigens.) It is only recently that serological techniques have been applied to detect allelic forms of proteins coded by these regions. In the mouse it has been shown that these gene products are also involved in graft rejection and cell interaction; between T cells and B cells for the production of antibody, between different populations of T cells to generate suppressor cells, and between macrophages and T cells. The other important immunological phenomena associated with the I region are the immune response genes (Ir genes) which control the response to specific antigens. Ir genes have been found associated with the MHC in all species in which they have been looked for but so far in man the evidence for them is more circumscribed, mainly because immunisation with the appropriate antigen would be unethical. We will first discuss the biochemical properties of the products of the genes from this region and then their different biological properties.

Use of antisera to study the I region

Antisera specific for I region antigens have revealed several different antigenic systems on mouse B lymphocytes⁸². The antigens are also found on macrophages, sperm, epidermal cells^{82,83} and on some T cells, particularly T cells which have been activated by con A (refs 84–86). They are notably absent from erythrocytes and platelets. The different antigen systems are coded by different subregions of the I region, I-A, I-C and I-E and so on^{82,87}. The alleles of each system are described by a series of numbers representing individual determinants and studies on wild mice suggest that there may be private and public specificities as found on the CHAGS⁸⁸. In the inbred strains of mice not all the specificities have yet been assigned to I subregions and new specificities and I subregions are still being discovered. The term Ia antigens refers to all antigens associated with the I region. It is important to stress that the molecules carrying the numbered specificities described above represent only a part of the Ia antigens. For want of a better term we shall call the numbered Ia specificities the B-Ia antigens.

Membrane molecules have been isolated which carry B-Ia antigenic determinants⁸⁹. The specificities coded by genes in the I-A and I-C subregions are carried on different molecules^{88,90,91}. On the other hand the specificities determined by one allele from either subregion have so far been found on the same molecule⁹². The molecules carrying the B-Ia specificities are glycoproteins each of which consists of two different polypeptide chains held together by disulphide bridge(s). It is not known if both chains carry antigenic determinants coded for by the I region. The molecules carrying the B-Ia determinants are not associated with β_2 -microglobulin on cell membranes.

There is a system similar to the mouse B-Ia antigens on human B lymphocytes^{93–97}. Genes coding for these antigens seem to lie at several loci, at least one around the HLA-A locus and a second to the left of the HLA-B locus. In fact

one serum seems to show complete concordance with HLA-D typing⁹⁸, though this is not generally the case. This work is still at an early stage and it is not yet known how many loci are involved or the extent of the polymorphism. As yet there is no unified terminology for this system of antigens and although it has not been conclusively shown that they are analogous to the mouse proteins we shall call them the human B-Ia antigens. The existence of strains of mice which differ only in the I region and are otherwise genetically identical, makes it relatively easy to define mouse Ia antigens. In humans the absence of such defined genetic differences makes it very difficult to be certain that any anti-B-Ia antisera do not also contain activity to other unrelated antigens. The molecules carrying the human B-Ia determinants have not been definitely identified but from knowledge of the mouse antigens obvious candidates are the glycoproteins which contain two polypeptide chains which can be extracted from human B-cell membranes^{99,100}.

I region and the mixed lymphocyte reaction (MLR)

As we have already mentioned, differences in the I region evoke a 'strong' MLR⁸². This is measured by following the stimulation of DNA synthesis, after mixing lymphocytes from two different strains or individuals¹⁰¹. The cells which respond and proliferate are T cells¹⁰². Other regions in the MHC can cause an MLR but the response is usually much smaller¹⁰³. (In the mouse the M locus unlinked to the MHC can also cause an MLR¹⁰⁴.) The cells which are particularly adept at stimulating a response in an MLR are B-lymphocytes. Other cells including T cells, sperm¹⁰ and possibly epidermal cells¹⁰⁵ can also stimulate an MLR. In the mouse anti-B-Ia antisera directed against the stimulating cells will readily block their capacity to stimulate an MLR¹⁰⁶. Although it is notoriously difficult to interpret such experiments, it is natural to suggest the molecules carrying the B-Ia specificities described in the previous section are the molecules which are involved in stimulating the T cells which respond in MLR.

In man differences at the HLA-D locus provoke a strong MLR^{107,108} which has been exploited to type cells for this locus^{97,109}. The HLA-D locus shows balanced polymorphism but as yet the extent of the polymorphism is not known. The idea that B-Ia antigens evoke an MLR is again supported by the fact that one anti-human B-Ia antiserum reacts only with cells typed as HLA-D-DW3 by MLR⁸⁸. More usually, however, B-Ia antisera react not only with all cells with a particular HLA-D allele, but also with some cells carrying different HLA-D alleles; and some sera show no concordance with HLA-D at all⁹⁷. This type of result is difficult to interpret as HLA-D typing by MLR and serological typing may well have different sensitivities, and further because there is increasing evidence for cross reactivity between different HLA-D alleles⁹⁷.

I region and cell cooperation

The interactions between various cells of the immune system seem to depend on the compatibility of the interacting cells at the I region. Until recently, however, it was not clear whether that was because I region products played an essential part in cell cooperation, or simply because of an allogeneic reaction which abrogated cooperation. By the use of several techniques for eliminating the problem of allogeneic reactions, it has recently become possible to clarify the issue and it seems that I region products play an essential part in cell-cell cooperation, but that I region compatibility is not always necessary.

For example, cooperation between helper T cells and B cells in the production of antibody does not occur when the T and B cells are incompatible at the I region^{110–112},

unless an allogeneic reaction is prevented by the use of tolerant T cells^{113,114} or T cells selected for their inability to respond to I-region differences¹¹⁵. In interactions between antigen, macrophages and T cells it seems that I region products may play a part in recognition¹¹⁶⁻¹¹⁸. Helper T cells will produce a secondary response to an antigen-macrophage complex only if the antigen is presented on a macrophage carrying the same I region products as those on the priming macrophage^{117,119}. (In this case the effect of histocompatibility reactions have not been fully investigated.) It is now possible to circumvent the complications arising from allogeneic reactions altogether by using soluble T-cell factors to study the control of the immune response.

These factors fall into two classes. First, there are antigen specific factors which bind to their corresponding antigens and lead to specific responses¹²⁰. Second, there are non-specific factors which are induced by antigens¹²¹, mitogens (O. Sjöberg, unpublished) or an MLR^{122,123}, which enhance an antibody response to certain antigens¹²⁴. The antigen specific helper factor is not classical antibody and at least part of it must be coded for by genes in the I region since it reacts with anti-Ia sera¹²⁵. Treatment of B cells with anti-Ia antiserum prevents the cells from absorbing factor, which implies that at least parts of the B-cell acceptor sites for the T-cell factor are also coded by genes in the I region¹²⁶. Thus two different, but interacting, products of the I region are involved in T-B-cell cooperation. Preliminary evidence suggests that the acceptor molecule carries the B-Ia determinants and it may be the acceptor molecules that are responsible for stimulating an MLR. All that is known of the antigen specific factor is that it is a glycoprotein of molecular weight about 50,000. So far no allogeneic barrier has been found for the helper factor¹²⁵ and indeed mouse factor will work with human lymphocytes¹²⁷.

A second similar, but not identical, antigen specific factor is involved in T cell-T-cell interactions in the generation of suppressor T cells¹²⁸⁻¹³¹. The factor binds to antigen, reacts with anti-Ia sera but not anti-immunoglobulin sera and has a molecular weight of approximately 50,000. The factor is absorbed by T cells but not B cells¹³². This suppressor factor shows certain allogeneic barriers in that factor from one strain of mice will not react with all other strains. It has been suggested that the ability to absorb the suppressor factor is also related to I region products which mean that the I region is involved in two systems of interactive gene products important in regulation of the immune response¹³². It is clear that suppressor and helper systems are different but it is quite possible that they share genetic elements.

These antigen specific factors are likely to be closely related if not identical to the actual receptors for antigen on T cells. It has so far proved extremely difficult to characterise unambiguously antigen receptors on T cells, particularly as T cells will absorb serum antibody^{133,134}. By using antisera which react with idiotypic determinants on the variable region of immunoglobulin heavy chains it is possible to obtain evidence that an antigen receptor on T cells contains at least parts of heavy chain variable regions^{135,136}. To date there is no evidence that the antigen specific factors described above contain any part of immunoglobulin molecules, including the variable regions. Other factors which may be important in the regulation of the immune response, do react with anti-immunoglobulin antisera^{137,138}. There is no simple way to explain these apparent contradictions concerning the nature of the T-cell receptor for antigen but it is clearly important to find out the exact chemical nature of these antigen specific factors.

There are several ways that lymphocytes can be provoked into producing a nonspecific factor which can enhance the response of B cells to particular antigens. One factor with this activity seems to be a complex of β_2 microglobulin and

Ia antigens^{123,139} though other functionally similar factors produced differently do not react with anti-Ia sera (ref. 140 and A. Schimpl and E. Wecker, personal communication). The relationships of these nonspecific factors to each other and to the specific factors are not known.

Immune response genes

The discovery of immune response (Ir) genes in the MHC is extremely important and has acted as the catalyst for much of the work on the I region. It is known that certain individuals within a species fail to respond to a specific antigen and this deficiency maps within the I region. So far, in different mouse strains, defects have been found in the response to more than 40 antigens. The Ir genes controlling these responses lie in all three subregions of the I region. In every case the absence of response is recessive—that is, response is dominant^{1,141-143}. Further analysis of these defects has revealed that for at least some of these antigens there exist at least two Ir genes which can complement each other^{126,144-149}. Thus in some cases the hybrids between two strains which do not respond to a particular antigen may themselves respond. The existence of complementation groups suggests that at least two types of I region product are necessary for an immune response. The antigen specific factor and its acceptor described in the previous section are obvious candidates as products of a complementing gene system and indeed with certain antigens there is some evidence to support them^{126,150,151}. Similarly it has been found that there are at least two complementation groups for the generation of suppressor T cells^{152,153}. I region products have been shown to be involved in the regulation of the response to a wide variety of antigens though Ir gene defects are only seen for antigens which carry a restricted number of determinants. This suggests that responses to complex antigens involve the products of several complementing systems. Defects in a component of one system need not affect the activities of the others.

There are many immune response genes which are not associated with the MHC, some of which have been mapped to particular loci. Recently it has been shown that the products of one of these other Ir-genes is important for the interaction of antigen specific helper factor with B cells¹⁵⁰.

The complement system and the MHC

There are genes in the MHC which affect the complement system, a series of serum proteins activated characteristically by antibody-antigen reactions. These genes may function two ways. First, as structural genes which code for certain of the complement components. The most convincing evidence for this comes from linkage studies of allelic variants of certain complement components. Second, there may be a gene or genes which control the level of certain complement components. The first indication that genes affecting the complement system lay within the MHC came from work on the mouse Ss-Slp system. Ss and Slp determinants are found on two serum proteins whose levels are affected by sex hormones and by genes in the H-2 region, now mapped between I and D^{1,81}. Anti-Ss antiserum was shown to inhibit complement mediated haemolytic activity of mouse serum¹⁵⁴ and it now seems that Ss protein is mouse C4 (refs 155 and 156).

In man and rhesus monkey several allelic forms of factor B can be detected by electrophoresis. The genes coding for these variants are linked to the MHC^{157,158}. The structural gene in man for factor B has been tentatively placed to the left of HLA-B¹⁵⁹. Since factors B and C2 have analogous properties in the alternative and classical complement pathways and very similar physicochemical characteristics it has been suggested that the structural genes for these proteins are tandem duplicates¹⁶⁰. Recent evidence suggests that the C2 electrophoretic variants are indeed

linked to the MHC¹⁶⁰. Structural genes of this type, lying within the MHC, may be useful for understanding the pressures which generate balanced polymorphism at other loci. If these pressures operate on the MHC as a whole one would expect to see balanced polymorphism in complement components coded for by MHC genes. This would only be true if such complement components were able to accommodate sufficient structural changes and remain functional. So far only four functional alleles of factor B have been found, two of them uncommon¹⁶¹.

In addition to the structural genes for complement components in the MHC there may be genes which control the serum levels of these components. The Ss-Slp system may contain this type of gene. Deficiencies of particular complement components could represent either deletion of structural genes or malfunction of a controlling gene. In man C2 deficiency is linked to HLA in a number of family studies and seems to be in linkage disequilibrium with the haplotype A10-BW18¹⁶²⁻¹⁶⁵. Deficiencies of C4 and C8 have also been found to be linked to particular HLA haplotypes although only one family has been studied in each case¹⁶⁶. If these deficiencies represent malfunction of genes controlling the levels of complement components then it is possible that only one such gene exists in the MHC and that this gene may also be responsible for the observed MHC influence on levels of the receptor for complement components on lymphocytes¹⁶⁷. Naturally not all the genes for the complement system lie within the MHC C3 and C6 electrophoretic variants in man^{168,169} and Clr (man)¹⁷⁰ and C5 (mouse)¹⁷¹ deficiencies are unlinked to the MHC.

MHC and disease

Many diseases in humans have an association with particular CHAGS¹⁷². These associations can be divided into four types, each of which may have a different origin.

(1) The only very strong association is that of HLA-BW27 with ankylosing spondylitis and a related group of diseases. About 95% of Caucasian patients with ankylosing spondylitis have this allele¹⁷³⁻¹⁷⁵. It is likely that the cause of this strong association lies in a property of the BW27 protein itself, particularly since it is found in other racial groups^{176,177}. There are various ways in which CHAGS might directly influence the susceptibility to disease. For example, they could function as receptors for viruses or they may resemble certain pathogens so closely that the animal is unresponsive to that pathogen through self-tolerance^{7,52}. The absence of BW27 in a few patients with ankylosing spondylitis is still compatible with the theory that the BW27 protein is directly involved in susceptibility to the disease. Ankylosing spondylitis is a multifactorial disease, and in the BW27 negative patients there may be an excessive accumulation of the other factors involved.

(2) There can be weaker, but still significant associations of diseases in the general population with individual CHAGS, for example thyrotoxicosis and some other autoimmune diseases with B8 (ref. 178) and psoriasis vulgaris with BW17 (ref. 179). A weak association would be expected if the genes responsible for increased susceptibility were closely linked to the CHAGS and remained with particular CHAG alleles through linkage disequilibrium. However, these diseases could be multifactorial and the CHAGS themselves may be one of the factors directly involved in increased susceptibility. In this case as with ankylosing spondylitis one would expect to see the same CHAG allele involved with the disease in studies of different races.

In general if immune response genes are the reason for the observed associations, an allele leading to an absence of response would only have a deleterious effect in homozygotes as response is dominant. On the other hand, alleles which lead to an excess, or normally unwanted, response

would be dominant in heterozygotes. The diseases in the categories discussed above are in general not found with greater frequencies in MHC homozygotes. Consequently non-responder alleles are unlikely to be responsible for disease association. Many of the diseases in these two categories have an auto-allergic element which would be expected if the deleterious Ir-genes lead to an autoallergic response.

(3) A third category of association of CHAGS with disease are those cases in which there is a higher frequency of particular CHAGS among individuals with greater resistance to particular conditions. For example, there is a higher than average proportion of HLA-A2 positive individuals among long term survivors of acute lymphocytic leukaemia¹⁸⁰. This association may again be due to a rare responder allele of an immune response gene linked to HLA-A2 but in this case the response is beneficial. Linkage to beneficial responder alleles could also be responsible for the observed low proportion of particular CHAGS among individuals suffering from certain conditions¹⁸¹ (for example HLA-BW15 and chronic viral hepatitis)¹⁸².

(4) Finally, it is possible to find a particular CHAG associated with a disease within a family though not in the general population. For example in ragweed allergy sensitivity is inherited in particular families together with an MHC haplotype¹⁸³. In this situation the gene involved is unlikely to be a CHAG gene but some linked gene, again probably an immune response gene.

In all cases if the association of disease to CHAGS is through linked Ir genes it is sensible to look for associations with other markers in the MHC. Several studies have used the HLA-D locus for this purpose^{93,184}. In many cases association with HLA-D is stronger than with CHAG. This would be expected if the important genes were the Ir genes providing the relative distribution of MLR and Ir genes is the same in man and mouse. However, there are many immune response genes and their exact distribution is not known, even in the mouse, so that diseases most strongly associated with the CHAG could in fact be explained by close linkage of an Ir-gene. It is also possible that several genes in the MHC influence susceptibility to a single disease. Under such circumstances one would expect to see the disease associated more strongly with a haplotype than with individual markers¹⁸⁵.

Finally, in a few cases the association of disease with the MHC is probably a consequence of defects in the complement genes. For example, systemic lupus erythematosus is found associated with C2 deficiency and hence with the haplotype A10-BW18¹⁸².

Coda

It should be clear from what we have written that there is still much to be learnt about the MHC and it is probably premature to attempt a rational synthesis of all the facts. Phenomena which need to be explained are the balanced polymorphism at some loci, linkage disequilibrium, and the biological function of the CHAGS and determinants responsible for an MLR. In addition, the implications of the maintenance of these immunologically important genes in one complex during evolution are not understood.

It is possible to construct a moderately coherent explanation of the facts with the products of the immune response genes playing a central role. As these genes are codominant and it is usually desirable to minimise the number of determinants to which an individual cannot respond, there will be heterozygous advantage. This is particularly true if the number of immune response genes approaches the limit for stable tandem duplication. This accounts for balanced polymorphism of immune response genes which could be transmitted to the CHAGS through linkage dis-

equilibrium, provided that CHAGS can accommodate many amino acid replacements without losing whatever biological function they have. The balanced polymorphism of the CHAGS might simply be a consequence of their linkage to the Ir genes and have no relationship to their own biological function.

The properties of the products of the I region could also account for linkage disequilibrium. The fact that products from separate immune response loci in the MHC have to react with each other to control the immune response would lead to a selective advantage to the transmission of pairs of alleles together as a haplotype.

The above synthesis does not explain why genes for complement components are found in the MHC, nor does it cope with the particular attention T cells seem to pay to the CHAGS in all cytotoxic reactions. An alternative explanation of polymorphism is that the CHAGS are involved in developing T-cell immunity to viruses and other pathogens¹⁸⁶. It has been shown that heterozygosity of the CHAGS on virus infected target cells increases the efficiency of T-cell cytotoxicity. There are many other ways by which pathogens can be recognised and dealt with *in vivo*, which will reduce the pressure for heterozygosity of the CHAGS. Although this model ascribes a biological function to the CHAGS it does not explain linkage disequilibrium or their association with Ir genes, nor is there at present any clear understanding of the mechanism by which antigens on virus-infected cells become associated with the CHAGS.

If the CHAGS act as the actual receptors for viruses, it is possible to give an alternative explanation for linkage disequilibrium¹⁸⁷. Any individual with a receptor for a particular viral pathogen will be at risk especially if that person does not carry the appropriate immune response gene, for resistance. Thus pairs of alleles will be encouraged to travel together—a CHAG allele and the appropriate immune response gene. Linkage disequilibrium in the intervening piece of DNA will ensue. There may be other structures in the MHC which act as virus receptors¹⁸⁸.

It is obviously important to know if the nature of the genes clustered together in the MHC is of some significance and has therefore been conserved in evolution. It is at present hard to imagine why particular genes are clustered together. It has been suggested that the MHC has evolved by tandem duplication from the *T/t* complex¹⁸⁹. The possible similarities between molecules from the *T/t* complex and H-2 complex lend weak support to this unlikely idea. It is obviously too early to say what relevance the *T/t* complex may have to the MHC and indeed it is really too early to promulgate any further unsubstantiated speculation.

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articles

Radio-echo layers and the recent stability of the West Antarctic ice sheet

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A new method for studying polar ice sheets confirms the generally accepted concepts of ice sheet flow and finds that a region near the ice crest of the West Antarctic ice sheet has been stable for ~ 30,000 yr.

A CHANGE in the mass or configuration of the large continental ice sheets could affect world sea level, alter the average albedo of the polar regions, and change the climate^{1,2}. To determine whether the ice sheets have changed, are changing, or are likely to change in an im-

portant way, and to allow the interpretation of palaeoclimatic results of studies of deep cores it is necessary to understand the history and dynamics of ice sheet flow.

One way to study the present-day balance of an ice sheet is to compare data on ice movement with surface mass-balance (new snow accumulation) data. This can be done by comparing measured changes in the elevation of markers in the surface with local surface mass balance^{3,4}, or by comparing the horizontal flow of ice with the rate of up-glacier replenishment by new snowfall⁵⁻⁹. In general, however, the interpretation of such studies is difficult because

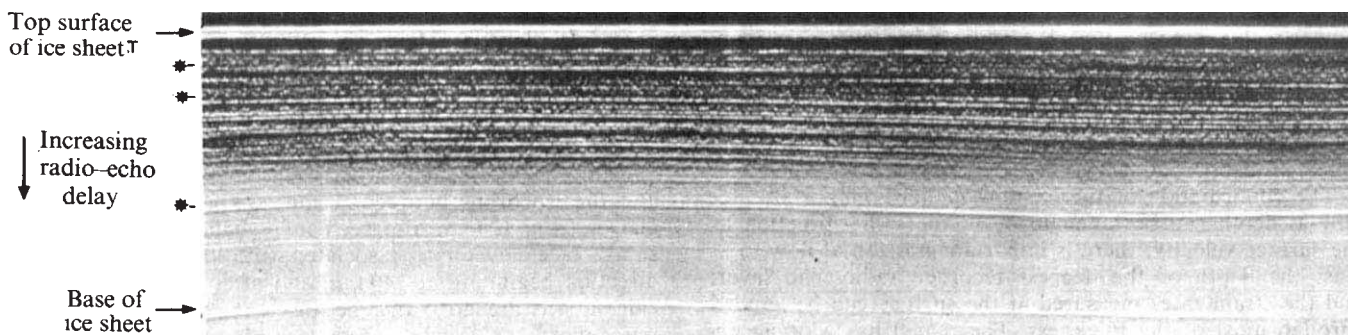


Fig. 1 Example of radio-echo record. A profile ~ 16 km long is represented. This example is located ~ 70 km from Byrd Station (Fig. 2), where the ice is $\sim 2,500$ m thick. Stars indicate the layers selected for analysis.

of uncertainties in measurement or because deep ice velocities are not known. A more satisfactory method for determining the health of an ice sheet is needed.

The recent histories of the Greenland and Antarctic ice sheets have been discussed using profiles of ice temperature¹⁰⁻¹², total gas content¹³⁻¹⁵, and ratios of the stable isotopes of hydrogen or oxygen¹⁵⁻¹⁸. Although very valuable, none of these techniques can distinguish clearly between changes in the form of the ice sheet and climatic changes.

A new approach is described here for studying the present and past dynamics of large ice masses. The method combines measurements of surface mass-balance, surface strain-rate, and the form of internal layering in the ice sheet determined by a radio-echo sounding technique.

Method

During the austral summer 1974-1975, radio-echo sounding flights were conducted in the Antarctic¹⁹. Equipment, designed and built by the *Danmarks Tekniske Højskole* (Lyngby, Denmark), and operated by members of the Scott Polar Research Institute (Cambridge, UK), was flown in an LC-130 (Hercules) aircraft by the US Navy as part of the United States Antarctic Research Program. Figure 1 is an example of a record from these flights: it shows radio reflections not only from the surface and base of the ice sheet, but also from within the ice mass. Many of these internal layers can be traced horizontally for >170 km. It has been suggested that the internal reflections are associated with sedimentary variations in density or impurity content, or old refrozen melt layers²⁰⁻²³, with certain annual layer spacings, or with horizons at which the mean ice crystal orientation changes²¹.

Flights were made along and perpendicular to the axis of the Byrd Station Strain Network (BSSN), which is up-glacier from Byrd Station, Antarctica (Fig. 2). Surface velocities and surface mass balances have already been measured along the BSSN^{9,24}, and using these data, it is possible to calculate the depths of ice layers that were deposited at different times in the past. The form of these buried layers is compared here with the internal layering revealed by the radio-echo sounding.

The model

A very simple model for the flow of the ice sheet is used. It is assumed that the strain rates, as measured at the surface, are constant throughout the thickness of the ice mass, but they vary in horizontal directions. The ice sheet is initially assumed to be in a steady state (with ice thicknesses and velocities independent of time), but possible departures from the steady state can also be discussed. The model

cannot adequately describe the flow of ice over and around variations in the bed, but is conceptually simple and adequate for present purposes.

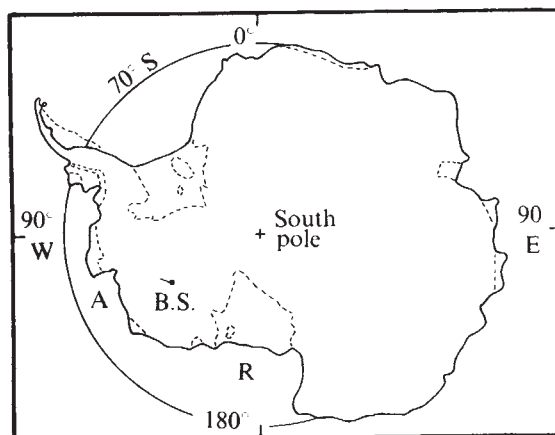
According to the model, the horizontal velocity of a vertical column of ice is the same at all depths, and variations in the bed of the ice sheet do not generate strains within the ice sheet except those that are measured at the surface. A layer of snow deposited at the surface, after compaction to ice, becomes buried by later accumulation, is thinned by the extending flow, and is carried down-glacier as the ice sheet flows towards the sea. The depth of a layer of any age may thus be calculated, and unlike the rather similar models of Robin and others²³, and Harrison²¹, ice thickness and bottom topography are not used in the computation.

The model results and the radio-echo results are independent and the model was developed before the radio sounding flights were conducted. The strain rate and velocity, as measured at the surface, are, in part, physically determined by ice thickness and bottom topography, so that some correlation between calculated buried layer form and the base of the ice sheet is to be expected for larger scale features of the relief.

Model—layering agreement

Figure 3 shows a profile of the ice sheet along the axis of the BSSN. The data for the depth calculations come only from surface measurements along the BSSN. The calculated isochrons and the measured radio-echo layering show agreement that is much too good to be coincidental, confirming the results of a similar study made along a flowline in northern Greenland²³. This shows that the simple ice-flow

Fig. 2 Antarctica, showing the location of the network (short straight line), Byrd Station (BS), and the Amundsen and Ross Seas (A and R).



model is a good approximation for the flow of the ice sheet, supports the concept that the radio-echo layers are connected with the depositional stratigraphy, confirms the conclusion that, at present, this part of the ice sheet is almost in a steady state⁹, and, finally, indicates that this portion of the ice sheet has been close to its present form for a long time: probably $>30,000$ yr.

The confirmation of the ice-flow model is important. Ice flow at depth can be taken to be in the same direction as the surface velocity; there is important horizontal flow to at least the depth of the deepest recorded radio-echo layer; and the strain rates measured at the surface can be applied through most of the thickness. The model has wide acceptance in the literature but there are few other tests of its validity for an ice sheet. Perhaps the best partial test so far arises from the measurement of the inclination change of the Byrd Station hole²⁵, which showed that the ice velocity vectors under Byrd Station lie close to a single plane, and that there are no sudden changes in shearing rate. This is for just one site, however, and the inclination changes could be measured only for the upper 1,490 m of the ice sheet.

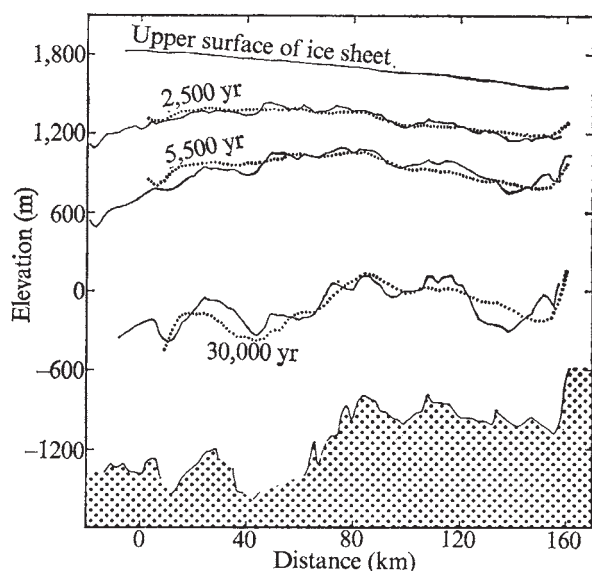


Fig. 3 Profile along the axis of the BSN. Three internal reflection layers and the base of the ice sheet have been obtained from the radio-echo records. The dotted lines show the calculated positions of ice layers of the ages specified. The ages have been selected so that the isochrons are at the same general depth as the selected radio-reflecting layers.

Analyses of the temperature distribution in ice sheets also find that a steady-state, uniform strain-rate model of flow similar to that used here can describe the measured temperature profiles. Changes in the glacier regime that may have occurred more than several thousand years ago may not, however, be detected by the temperature analyses, and differences in flow direction or vertical strain rate between the upper and lower portions of the ice mass may not greatly affect the agreement of the calculated and the measured temperature profiles.

The success of the model is important to studies that have been conducted with data from the deep core hole at Byrd Station. The ice-flow model used in the analysis of the temperature profile¹¹ and the depth-age relationship for the ice core as calculated by some workers^{17,18} are confirmed. The depth-age relationship obtained from microparticle layering²⁶ is not, however, consistent with the results presented here. Perhaps the microparticle layering is not annual^{27,28}, as was assumed in that work.

The radio-echo layering and the isochrons do not agree exactly. To a large extent this may be because the strain network and the flight lines are in slightly different positions. The depth of the layering is variable in the third dimension, as shown by the results of flights perpendicular to the BSN axis, and a small navigational error could explain the discrepancies between the radio-echo layering and the isochrons between 0 and 40 km in Fig. 3. Down-glacier from 120 km disagreement is believed to arise from sub-surface advection⁹ associated with the change in flow direction beginning at 140 km, and after 140 km the model calculations are not valid, because the strain network does not follow the flowline. Smaller differences between the model and the radio-echo layering are associated with irregularities in the bed, a factor that is not adequately treated by our simple model.

Each radio-echo layer is probably not caused by a single physical surface within the ice sheet^{21,22}. Sufficiently large changes in permittivity or loss tangent are probably too infrequent to cause all the layering in Fig. 1. More likely, the combined effect of many small changes in permittivity or loss tangent within the ice sheet causes each single layer on the radio-echo record. Each radio-echo layer may be associated with material injected into the atmosphere, perhaps from a volcanic eruption, and the material was for many years seasonally deposited on the ice sheet according to seasonal shifts in weather patterns²⁹⁻³².

Other explanations for the success of the model?

We suggest that the radio-echo layering is associated with stratigraphic layers of certain ages, but it could also be argued that the radio returns are associated with certain spacings in stratigraphy²¹; for example, annual layering. This spacing would be related to the wavelength of the radio wave in ice, such that reflections from all, or many, of the annual layers would be in phase, and therefore reinforce each other. Using the simple steady-state model for ice flow described above, the depth at which the annual spacing is equal to certain selected values has been calculated and is shown in Fig. 4. The upper two radio-echo layers do not correlate well with these contours and the possibility that only certain annual layer spacings cause the radio reflections is discounted.

Except for the probably small influence of bottom melting or freezing, the oldest isochrons and the thinnest annual layer spacings should both be near the base of the ice sheet, so that very old isochrons and contours of very thin annual layer spacing should be similar, and the fair agreement of the 0.03-m contour in Fig. 4 with the deep radio-echo layer is expected.

It may also be argued that the radio-echo layering is caused by ice foliation²¹ from shearing flow in the ice sheet. This is plausible because bands of different crystal size and crystal orientation are reported for the Byrd Station core³³. Foliation, however, does not become well developed until deeper than 1,250 m below the surface (300 m a.s.l.) at Byrd Station (at 162 km in the Figs)³³ and the radio-echo layering is well developed above this level (Fig. 1). Furthermore, near-horizontal foliation is not likely to have been strongly developed in the ice crest region where horizontal shear strains are orders of magnitude less than elsewhere, yet the radio-echo layering continues, uninterrupted, across the ice crest (at 0 km in Figs 3 and 4). The simplest explanation for the radio-echo layering is that they are isochrons.

Values for surface mass-balances and strain-rates other than those that have been measured could be used to calculate the isochrons. Agreement between the isochrons and the radio-echo layering would be equally good for values of surface mass-balance and strain rate that retain a constant proportion to the measured values (although the calculated ages of the layers at any depth would differ).

There are two distinct surface mass-balance regimes along the BSSN: the first, near the ice crest extending to ~40 km, has a larger surface mass-balance that decreases smoothly with distance from the Amundsen Sea; and the second region, the remaining 120 km, has a smaller surface mass-balance that varies according to the surface slope²⁴. The surface mass-balance is therefore determined by processes acting with different effectiveness in the two regions, so that the climate is unlikely to have changed in such a fashion as to affect both surface mass-balance regions in the same proportion. For example, a change in the frequency of snow-bearing storms from the Amundsen Sea would be expected to affect the surface mass-balance of the ice crest region more than elsewhere along the BSSN. This would change the relative depths of each isochron under the two surface mass-balance regimes and result in a poor correlation with the radio-echo layering.

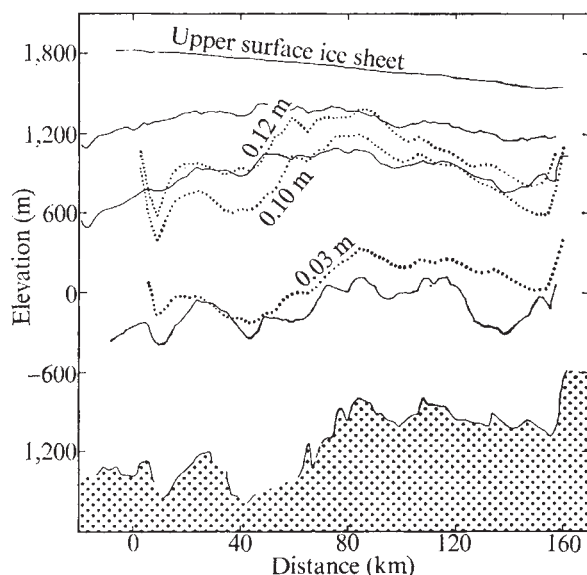


Fig. 4 Profile along the axis of the BSSN. As in Fig. 3, the radio-echo layering is shown by solid lines, but the dotted lines are the calculated positions of ice with the specified annual layer spacing. This is a test of the hypothesis that the observed radio-echo layering may be caused predominantly by the constructive interference of many weak reflections from certain regular spacings in the annual stratigraphy.

A uniform thickening or thinning of the ice sheet with time would change the mean strain rates in a constant proportion, but it is difficult to conceive a mechanism that would thicken or thin the ice sheet uniformly and significantly. Changes in surface mass-balance or air temperature would cool or warm the ice mass at different rates and by different amounts in different parts of the ice sheet, and thus cause important changes in flow lines and dimensions of the ice mass. Similarly, shifts in the position of the ice crest, caused perhaps by changes in the grounding zone, where the ice sheet begins to float in the Amundsen Sea or Ross Sea embayments, would affect the surface mass-balance distribution, and the flow directions and consequently the positions of the isochrons in the ice sheet. Changes at the seaward edge of the ice sheet would affect different drainage basins differently and cause shifts in the position of the ice crest: such a shift would have disturbed the layering of Fig. 3. The radio-echo layering in the BSSN region is consistent with the present-day flow pattern and surface mass-balance distribution and it is very improbable that a major change in flow or precipitation pattern could have resulted in internal layering that so closely follows present-day steady-state isochrons.

Ice sheet stability

The simplest and most plausible explanation for the simi-

larity of the calculated isochrons and the radio-echo layering is that the mass-balance, velocity field, and shape of the central portion of the ice sheet have not changed significantly during the past ~30,000 yr. Small changes in the surface mass-balance or strain rate may not be detected by the technique. The correlation of Fig. 3 is almost equally good using surface data locally changed by as much as ~5% from present-day values.

Other workers have suggested that the West Antarctic ice sheet is behaving unstably³⁴⁻³⁶, but we find that there have been no important changes in the past 30,000 yr. Peripheral parts of the ice sheet could, however, have recently begun retreating rapidly^{37,38}, or have begun advancing rapidly⁸, without the effect having yet reached the region of the ice crest, and we cannot predict future behaviour. The significance of the stability conclusion extends beyond the immediate BSSN region because distant thickness changes would have altered flow directions in the BSSN region and would appear as anomalies in the radio-echo layering. It is difficult to estimate over what area the steady-state result is applicable, but it probably extends 300 km from the BSSN, or over most of Marie Bryd Land.

The technique described here could be applied to other areas where the ice flow can be simply interpreted and where internal radio-reflections can be obtained. It should be used to help select sites for deep drilling and aid in the interpretation of deep core data, and, as shown here, it can be used to determine the balance at present and in the past (since ~30,000 yr BP, in this case) for a large ice sheet.

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Alternating subduction direction and the evolution of the Atlantic Caledonides

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The Morarian, Celtic, Grampian and Lakelandian orogenies are the result of alternating subduction north-westwards and then south-eastwards successively from 1,000 to 450 Myr at the margins of one major ocean basin. Subduction on both margins between 450 and 350 Myr caused the final closure of the basin and the Caledonian orogeny.

THE late Precambrian and lower Palaeozoic history of the north Atlantic region has been difficult to unravel in terms of plate tectonics largely because of the large number of apparent orogenic events and their differing ages in different segments. Most attempts have followed Wilson¹ by starting the history of the Caledonian system with an assumed phase of continental splitting to form the Iapetus² ocean at the beginning of the Cambrian. This separated north-west Scotland, north-west Ireland, and western Newfoundland from England, Wales, south-east Ireland, and E Newfoundland. Such an ocean must have existed at least from Cambrian times, as has been demonstrated³ on palaeontological grounds, but when the late Precambrian history of the region is studied it becomes very difficult to find unequivocal evidence of initial continental rifting. Alkaline activity at 565 Myr in the St Lawrence and Oslo Graben has been suggested⁴ as indicative of the initial rifting of the continental mass. Early Cambrian fossil assemblages from the American and Welsh areas, however, represent highly contrasted benthic faunas which presumably lay on opposite sides of a well developed ocean^{3,5} so it seems unlikely that the time of initial opening was as late as 565 Myr. The most significant feature of the ocean, however, that is difficult to reconcile with a repeated closing and opening, is the independent nature of the plate tectonic events taking place on either margin from 1,000 to 350 Myr.

I suggest here that Iapetus was an ocean of long standing. The successive climactic episodes of orogenic activity (deformation, metamorphism and some types of volcanic and plutonic activity), although nowhere precisely dated, do seem to fall into a remarkably consistent pattern of alternating activity on the north-west and south-east margins. Iapetus presumably had a spreading ridge along its axis, and I suggest that subduction took place alternately to the north-west and south-east, the subduction stopping after a climatic 'orogenic' phase and then transferring to the far side of the ocean. The known pattern of orogenies are thus produced in alternating sequence.

The cause of each orogenic episode then becomes the principal problem. It has recently been recognised that marginal basins may be important in the plate-tectonic evolution of orogenic belts and the Burlingtonian orogeny in Newfoundland has been ascribed⁶ to the closing of a marginal basin. The sequence of events recognised⁷ in the Andean chain of southern Chile seems a likely model. There a cordilleran orogenic belt split behind the subduction zone and a series of marginal basins of oceanic material formed behind a continental sliver. Subsequent closure of those basins was accompanied by considerable compressional

deformation and the subduction in that area probably ceased soon afterwards. The reasons for the marginal basin closure and cessation of subduction have not, so far, been deduced.

A model for the development of the orogenic phases is suggested here (Fig. 1a-d) whereby subduction under a continental edge may lead to the development of a marginal basin beyond the volcanic arc/continental margin which eventually closes and causes an arc/continent collision orogeny. The subduction direction then changes to the far side of the major ocean. This produces a pattern of alternating activity on either margin, which is illustrated for the Caledonian of the North Atlantic by a series of five cartoons which portray the situation immediately preceding each of the following five orogenic events:

- (a) The Morarian⁸ before ~ 1,000 Myr.
- (b) The Celtic⁹ and Ganderian⁶ at ~ 600 Myr.
- (c) The Grampian¹⁰ and Burlingtonian⁶; Tremadoc-Arenig in age.
- (d) The Lakelandian¹⁰ somewhat similar in age to the Taconic of the Appalachians; Upper Ordovician in age.
- (e) The Caledonian or Cymrian¹⁰ orogeny; end Silurian to mid-Devonian from Scandinavia to Newfoundland. This marks the final closure of Iapetus.

The Morarian orogeny

The earliest evidence of continental margin activity on the site of Iapetus comes from north-west Scotland (Fig. 2). The deposition of the Stoer Group¹¹ at ~ 1,000 Myr may mark a post-orogenic molasse deposition to the Morarian orogeny. The Moinian¹¹ lay to the south-east and is a series of trough greywackes, the current markers indicating currents running north-west-south-east. In places it can be seen lying directly on Lewisian basement. It was involved in one or perhaps two periods of deformation¹² and the climax of metamorphism produced a migmatitic core at 1,050 Myr (ref. 13). Later pegmatites were intruded at ~ 730 Myr (ref. 14). The folds include large scale nappes whose direction of transport was probably towards the south-east, and the grade of metamorphism and the ductile deformation of both the crystalline Lewisian and the Moinian sediments suggests that there was a collision orogeny in this region ~ 1,050 Myr ago. This has been termed the Morarian⁸.

A similar event has been recognised in the Ox Mountains inlier of north-west Ireland^{15,16}. The Deer Park Complex of Clare Island has associated amphibolites and serpentinites which continue to the east and north into the Ox Mountains area¹⁶. They are considered to be older than the upper Dalradian and may thus represent part of a Morarian Suture Line. They lie between the possible continuation of the Great Glen Fault and the Highland Boundary Fault—possibly in part even to the south-east of the latter. It thus seems from this evidence that the Morarian metamorphic belt extended south-east of the Great Glen and forms the basement to the Dalradian Supergroup.

It is thus suggested (Fig. 2) that the Moinian developed on the continental margin behind an island arc, and was deposited some way landward of the subduction zone and

any associated igneous activity. The collision of the island arc with the margin of the continent is envisaged as the cause of the Morarian orogeny.

Elsewhere in the North Atlantic region this was the time of the Grenvillian and Riphean orogenies. Both of these areas are characterised by the apparent lack of an extensive depositional episode and the high grade, in places granulite

facies, metamorphism of early Proterozoic rocks. Anorthosite plutons seem characteristic of this belt and late orogenic granitic bodies as late as 850 Myr occur in Telemark¹⁷ and Sweden. The Grenville front, which probably extends across the Rockall Bank¹⁸, is approximately at right angles to the Riphean front on Bullard, Everett and Smith's reconstruction of the North Atlantic¹⁹. Certainly neither the Grenville nor Riphean fronts represents a suture line, though there may be such within the central parts of these complexes. Thus, although the recent determination of a 'Grenvillian' age for the Morarian high grade metamorphism¹³ is indicative of orogenic activity at the same time as the Grenville and Riphean events it seems unlikely that the Morarian represents a simple continuation of the Grenville to link it with the Riphean. It may be that south-eastern Scandinavia was at this time on the northerly side of Iapetus (Fig. 2) and that the ocean passed south-eastwards into mid-Europe between Scandinavia and the Low Countries, but evidence is lacking in this critical area. In any case the position of the Morarian seems rather anomalous regarding the amount of sedimentation and the type of deformation. Perhaps the Scottish and Irish areas represented a small back-arc basin to the main Grenvillian-Riphean orogenic belt which elsewhere developed as a Cordilleran orogeny largely by reworking of continental crustal material and without a significant sedimentary prism.

The Celtic-Ganderian orogeny

Subduction then switched to the SE margin of Iapetus (Fig. 3) and probably at this time also the Scandinavian arm of Iapetus was extended NE from Scotland as shown by Precambrian sedimentation in E Greenland through to N Norway. The trough sediments of the Mona and Cullinstown Groups were almost certainly laid down adjacent to, or upon, a continental crustal segment possibly of Archaean age. But they were also near oceanic crust as Anglesey contains most of the indicators of a subduction zone: greynwacks intruded by a serpentinite-gabbro diapir, wildflysch, pillow lavas, cherts and basic rocks raised to blueschist facies. The upper part of the sequence contains acid

Fig. 2 Formation of Morarian (> 750 Myr).

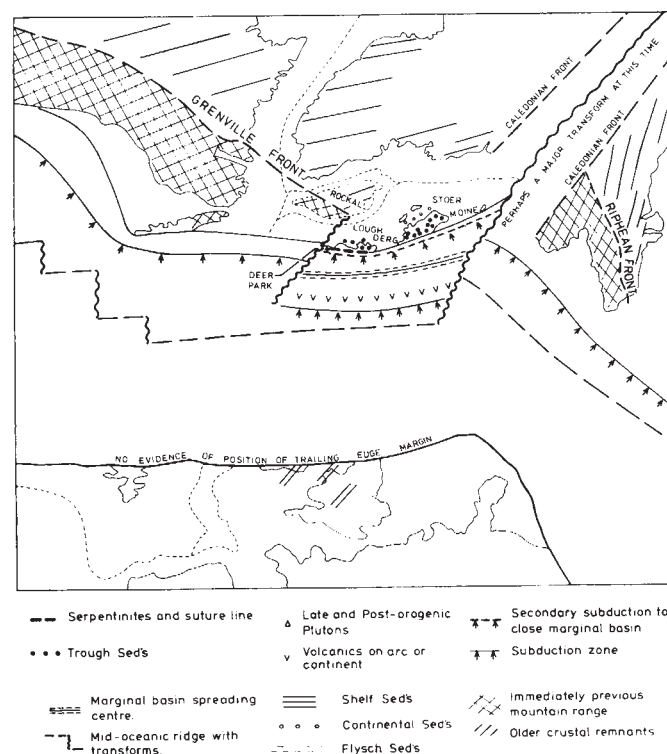


Fig. 1 *a*, Subduction with trench and trough sedimentation (*A*) leads to development of volcanic arc (*B*) either on continental crust thinned by the initial rifting (as shown) or on oceanic crust. Some time after the initiation a marginal basin (*C*) forms behind the volcanic arc and continental margin shelf sediment develops on crust thinned by marginal basin formation (*D*). *b*, Arrival of hot light crust, *A* (mid-ocean ridge) at subduction zone (*B*). This is too light to subduce. Provided that the marginal basin is not too young (that is, its older parts are relatively cool) subduction is initiated within the marginal basin either at *C* or *D*. Sediments at *C* (or *D*) become trough sediments and may be raised to blueschist facies. Cessation of the main subduction causes the marginal basin spreading centre to become a dead ridge (*E*). By this time the far side of the ocean (*F*) has a thick shelf sequence on the trailing edge of the continent. *c*, Subduction within the marginal basin causes collision of the volcanic arc with continent at *A* with nappes on to continent (*B*) and probably on to arc (*C*). Ophiolite (of marginal basin type) and blueschist fragments may be caught in suture zone (*D*). Original trench zone (*E*) now becomes site of shelf sedimentation as this is now a trailing edge, the mid-oceanic ridge (*F*) now moving away from this continent toward the subduction zone (*G*) now on the far side of the main ocean. *d*, After the stopping of subduction and orogenesis at *A* (by a sequence similar to *a-c* above) subduction again starts at original side (*B*) and a new marginal basin (*C*) would be initiated in the thinner crust of the original volcanic arc which has been rapidly eroded and covered with shelf sediments (*D*). The new volcanic arc (*E*) may be partly on the older arc or further oceanwards, but is likely to be a good deal nearer the ocean than the original orogenic collision zone (*F*).

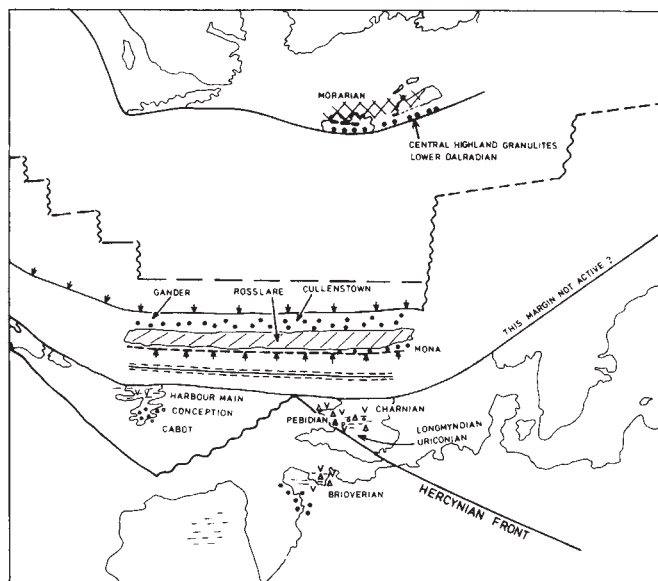


Fig. 3 Formation of Celtic and Ganderian (750–600 Myr). Key as for Fig. 2.

and intermediate volcanics indicating that in the later stages a volcanic arc supplied detritus to the trough area. To the south-east there was a widespread development of calc-alkaline volcanism and plutonism, all probably within the range 750–600 Myr, developed as a fairly typical cordilleran sequence on continental crust.

There seems to be no argument for a longstanding north-west dipping subduction zone since all the contemporary calc-alkaline volcanics lie to the south-east. It is more likely that a marginal basin developed between Anglesey and the Welsh Border and that a collision, closing this basin at ~ 600 Myr, caused the folding in Anglesey, and brought the blueschists and oceanic crustal slivers to the surface. The evolution of the belt in the English Midlands and Welsh borders follows the normal development of a cordilleran orogeny some way from a continental edge⁹.

In Newfoundland, Kennedy⁶ suggests that a Ganderian orogeny took place on this margin during the latest Precambrian, and advocates the same mechanism, although evidence for a marginal basin is lacking. The Cadomian¹¹ of Brittany and the Channel Isles has been more accurately dated than the rest of this area. It has been suggested²⁰ that Brioverian sediments developed on a south to south-eastward facing continental rise with possibly an ocean indicated by ophiolites in the south. This subsequently became deformed before 620 Myr by east-north-east folding and metamorphism which reached quite high grades in northern Brittany and culminated in the emplacement of late-orogenic intrusives as late as the Lower Cambrian¹¹.

The closely similar timing of the orogenic events of the Cadomian and the Celtic cycles suggest that they were in some way related but their relative positions at this time are unknown. It is possible that the relative positions of these two areas have been markedly altered during the Hercynian orogeny, although Ordovician fossil assemblages point to a single province in the whole of this area to the south-east of Iapetus²¹. On balance it seems most likely that the Brittany area was on a different continental margin. The late Precambrian of north-west Spain which has been compared with the Brioverian was not involved in any orogenic activity at this time²⁰.

The climax of the Celtic orogeny resulted in the formation of a mountainous landmass on the south-east margin of

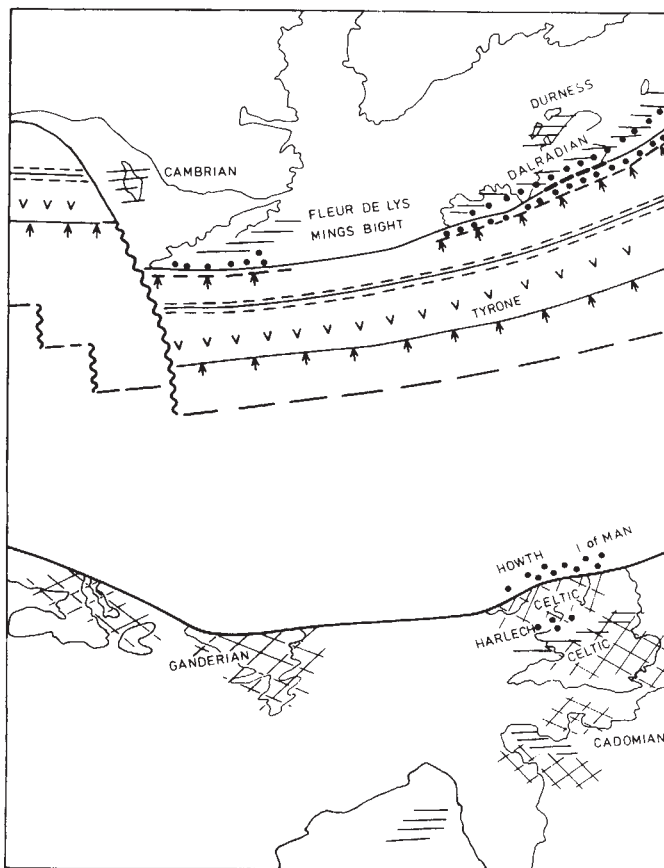
Iapetus and shortly after this there is evidence of increased activity on the north-west side of the ocean.

The Grampian-Burlingtonian orogeny

Following the formation of the Morarian mountains the north-west continental margin continued to receive trough sediments to the south-east of the Great Glen as the Central Highland Granulites (Fig. 4). These sediments show no earlier structural history than that affecting the Dalradian, so must be regarded as the lower part of that sedimentary sequence⁸. Similarly in western Mayo the Erris Group seems to be Moinian type sediment unaffected by any pre-Dalradian deformation¹⁵ and may again represent a post-Morarian early-Grampian trough.

Shelf sedimentation began in the lower Dalradian¹¹ and continued through Middle Dalradian (probably about the end of the Precambrian). All these sediments were being laid down, according to the present model, on the subsiding Morarian volcanic arc. From early Cambrian this shelf extended over on to the Torridonian beyond the Morarian chain, now largely eroded. In upper Dalradian (about Cambrian–Arenig), the continental margin began actively subsiding and great thicknesses of greywackes were deposited, probably marking the beginning of subduction to the north-west again. There are abundant basic volcanics (Tayvallich) in the upper Dalradian but they cannot represent slices of oceanic crust since they lie within a sequence of coarse, shallow-water quartzites. They could, however, mark the opening stage of a marginal basin. The intrusion of the Aberdeenshire layered gabbros (again, not a suture line) apparently after some of the early folding but before the climax of the orogeny indicates proximity to a source of abundant tholeiitic magma, and marginal basin activity would again seem a fairly logical source.

Fig. 4 Formation of Grampian and Burlingtonian. See Fig. 2 for key.



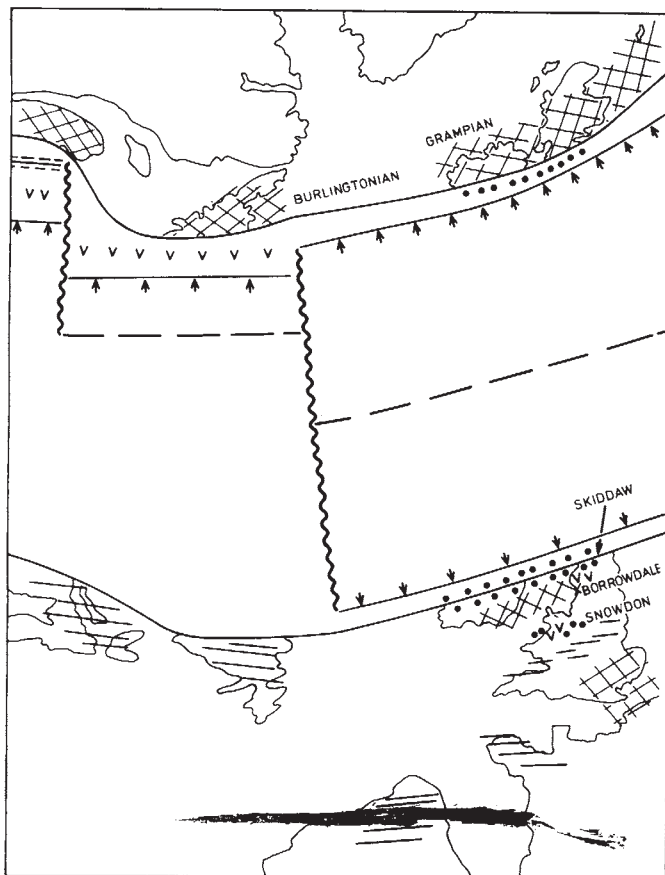


Fig. 5 Formation of Taconic, development of Lakelandian cordillera. See Fig. 2 for key.

The climax of the Grampian orogeny has not been precisely dated but must be about Arenig times. There was intense deformation accompanied by high temperature/high pressure metamorphism, so there is little doubt that it was a collision orogeny. A similar orogeny with clearer evidence of marginal basinal effects has been proposed⁶ for the Burlington peninsula of Newfoundland. This Burlington orogeny took place during the Tremadocian.

The necessity for a substantial collision to cause the Grampian orogeny without closing the whole Iapetus ocean (and thus involving the Lake District and Wales in a major Arenigian deformation), necessitates the suggested development of an island arc to the south-east of the main part of the Dalriadan. This would also explain the lack of calc-alkaline volcanics within the Dalriadan sequence, or indeed of volcanics of Cambrian age anywhere in the British Caledonides. The abundant plutonic basic and acid intrusions in the Connemara Dalriadan which were intruded at various times in the orogenic cycle could also be indicative of the proximity of that area to the major volcanic arc.

The cryptic suture which does exist along the Highland border (some Arenigian pillow lavas and serpentinite intrusions) probably thus only marks the line of closure of the marginal basin; the major continental fragment which may have had Cambrian calc-alkaline volcanic activity would now lie beneath and south-east of the Midland Valley. The recent LISPB experiment has shown that oceanic crust does not exist beneath either the Southern Uplands or the Midland Valley²³.

The Lakelandian and Taconic orogenies

Following the Grampian orogeny, subduction again switched to the south-east side with the development of middle-upper Ordovician volcanics and trough sediments in the Lake

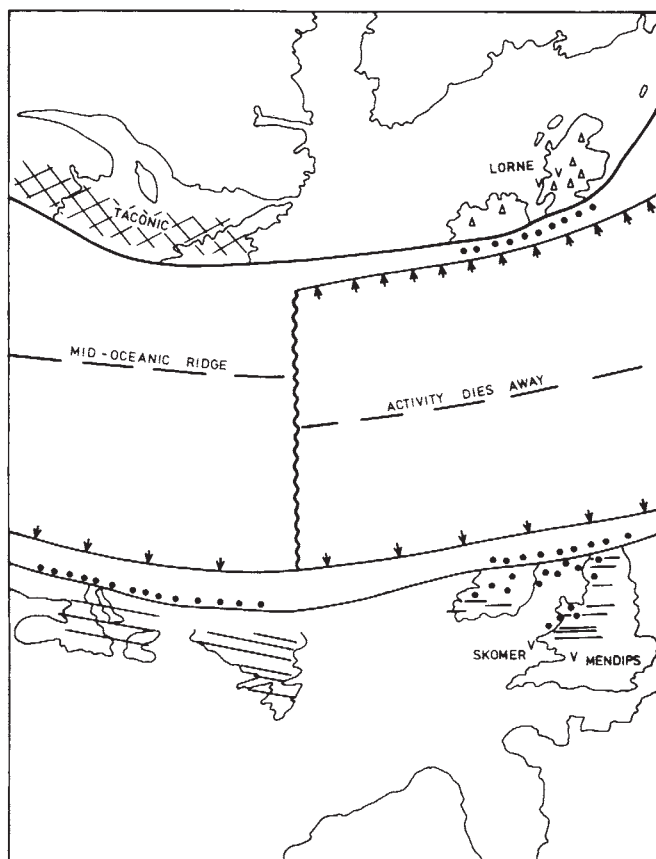
District and Wales (Fig. 5). This followed a period of basinal sedimentation between the Celtic trough area of Anglesey and the Celtic foreland of the Midlands which were both land areas for much of Cambrian and Ordovician times. Although black shales were at times being deposited, the mid-Welsh trough does not seem likely to have been a true oceanic area, but could represent an area of crustal thinning which never quite developed into a marginal basin (for example, the Llanvirnian Strumble Head Pillow Lavas could mark the opening of such a basin).

There are pre-Borrowdale and pre-Bala earth movements which have been termed the Lakelandian¹⁰ and regarded by some as a major orogenic phase²⁴. It is, however, obviously not a major collision and must be regarded as a cordilleran orogeny on this side of the ocean which never developed a marginal basin or resulted in a major collision orogeny.

On the north-west side of Iapetus (south of the Scottish Highlands) subduction may never have ceased after the Grampian orogeny, since oceanic crust of Arenig to Llandeilo age is preserved in the Girvan area²⁵ indicating either oceanic subduction or marginal basin activity in that area very shortly after the Grampian orogeny. In Newfoundland also, tectonic activity in Caradocian times emplaced large slivers of oceanic crust and mantle right over the Burlingtonian core⁶ and indicates that subduction to the north-west continued without much break after the Burlingtonian orogeny in the Newfoundland segment.

Further south-west in the Appalachians, the Taconic orogeny represents a major collision that does not correlate in timing or intensity with the Lakelandian. The age of the Taconic climax of activity is about Caradocian²⁶ and involves major thrusts and nappe type structures which, as with the Grampian, cannot be related to the Ordovician

Fig. 6 Closure of ocean. Formation of Caledonian and Acadian at final collision. Key as in Fig. 2.



history of the south-eastern side of Iapetus. This again implies that the two sides of the ocean were acting independently and the nature of the Taconic orogeny implies a major subduction and collision event in the south-western part of this belt during early and mid-Ordovician times on the north-western margin. It is perhaps most probable that the Burlingtonian orogeny did not take place there but was delayed until Caradocian times in the area south-west of Newfoundland.

The Caledonian orogeny

Whether subduction ceased to the south-east or not (volcanism continued to be active in Silurian times in the Mendips and South Wales), the calc-alkaline volcanism of Lorne and Glencoe suggest that active subduction was taking place beneath the Dalradian, with the subduction zone possibly in the Solway area²⁷ (Fig. 6). It may have been the effect of subduction to both north and south that allowed the closure of the Iapetus ocean, as by the middle of Silurian times the palaeontological evidence indicates that benthic fauna were in contact and by late Silurian or early Devonian the collision had taken place, marine sedimentation had ceased and the rocks of Wales, the Lake District, the Southern Uplands and Central Ireland became quite strongly folded. This is the first time that deformation at approximately the same time can be recognised occurring on both sides of Iapetus, another good indication that this was the first oceanic closure. Along the length of the orogenic belt the intensity of deformation associated with the major oceanic closure increases from Newfoundland (where it was mid-Devonian and very mild) to Scandinavia where it was much more intense than in Britain. In Scandinavia the Caledonian orogeny was so intense that Silurian rocks are metamorphosed and involved in major thrusting, so that the correlation of the orogenic events in that segment with the sequence proposed here has not been attempted.

This sequence of orogenic climatic events is thus envisaged not as a continuous orogenic cycle operating along a continental collision-site as many previous syntheses have portrayed the Caledonides, but as a discontinuous series of subduction/marginal basin formation/closure-collision events on either side of a long-standing major ocean which only finally closed in Silurian to mid-Devonian times and thus stopped any subduction or ocean spreading in this region. The model is clearly susceptible to testing by palaeomagnetic determination and may need revision in the light of local stratigraphic detail. The model does, however,

draw attention to one of the major features of plate tectonics applied to older palaeogeographic reconstructions that has not been too clearly recognised by many writers, namely the independence of events on one side of an ocean from contemporaneous events on the other. The correlations between the sedimentary history of the Mona complex with possibly contemporaneous sediments in the Dalradian or Brioverian will be practically impossible, and largely irrelevant, if these three areas were on three different continental margins at different stages of activity or of different geotectonic type. Similarly, the search for the equivalents of the Taconic orogeny in southern Britain are irrelevant if the two areas were on margins of two different, independently acting, continents.

A major problem now apparent from the series of suggested events (Figs 2–6) is the reason for the closure of Iapetus and specifically why its mid-oceanic ridge stopped spreading. One must look again to a coupled series of events and the beginning of the Hercynian orogeny is closely related to the end of the Caledonian, so that it seems very likely that the starting of a major site of crustal extension and the formation of an ocean on the margins of which the Hercynian orogenies developed²⁸, was the cause of the failure of the Iapetus spreading centre and ultimately the closure of that ocean forever.

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letters to nature

Oblateness of the atmosphere of Mars

ON April 8, 1976 Mars occulted the bright star ϵ Geminorum (magnitude 3.2). Predictions¹ were issued well in advance since such occurrences are very rare: indeed, it is estimated that Mars occults a star as bright as this about once in 500 yr. Accurate observations of the duration of the occultation were made from four sites on the Earth and these are analysed here for the shape of the Martian atmosphere at that height at which the intensity of the starlight diminished by half. The oblateness is found to be about twice as great as that of the surface of Mars.

The occultation was widely observed throughout North America, the Caribbean area and Great Britain, mainly visually, although a number of photoelectric observations were also made. Of particular interest are the high accuracy photoelectric observations made from Greenbelt (Maryland), Atlanta (Georgia), Fort Davis (Texas) (G. de Vaucouleurs, et al., to be published) and from an aircraft over the western North Atlantic². Timed observations of '0.5 light' at the disappearance and reappearance phases of the occultation have been reported from these four places. The aircraft observations, from a height of > 12 km, were made at 4,500 Å—a shorter wavelength than that used by the ground

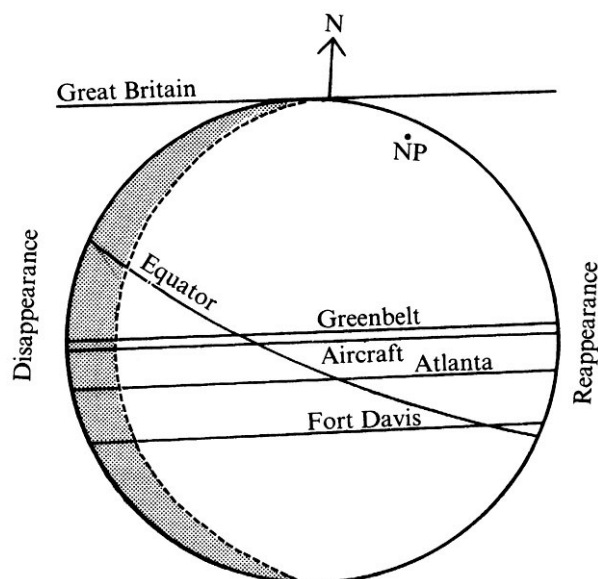


Fig. 1 Occultation of ϵ Geminorum by Mars on April 8, 1976 as seen by various stations. NP marks the North pole. Although the chords representing the photoelectric observations are nearly central, there is a good spread in latitude because of the tilt of the axis of Mars.

observers. Because of the very low altitude of Mars it was not possible to make photoelectric observations from the British Isles. This was unfortunate since a grazing occultation occurred from southern England, Wales and Ireland, and observations from localities such as these would have improved the coverage of the Martian disk considerably. This is clear from Fig. 1.

Using the known distance of Mars and its direction and angular rate of motion, it was possible to calculate the apparent positions of the star at the observed times, relative to the centre of Mars. A standard computer program, written by the author, for the analysis of previous occultations of stars by planets and satellites was used to solve for three unknowns—two for the position of Mars and one for the correction to the adopted equatorial radius. Several solutions were found using various trial values for the flattening of the Martian atmosphere, where the flattening is defined as [(equatorial radius—polar radius)/equatorial radius]. Calculations were also done without the aircraft observations since these were made at a shorter wavelength than the others and thus refer to a greater height in the Martian atmosphere. This did, indeed, prove to be the case, the height being ~ 6 km above the ellipsoid fitted to the other observations.

The position of the star was taken from the FK4 catalogue, while the ephemeris of Mars was obtained from the Jet Propulsion Laboratory. The analysis of the ground-based photoelectric observations showed that this ephemeris was extremely accurate. In fact, the derived correction to the ephemeris of Mars relative to the star was only

$$+0.0019 \pm 0.0001 \text{ s in right ascension (RA)}$$

$$+0''.009 \pm 0''.007 \text{ in declination (dec.)}$$

This correction is considerably less than the s.e. in the position of the star itself.

For the first solution the adopted value of the flattening was 0.0052, which is the dynamical flattening given for the surface of Mars by Lorell³. By varying this value, however, it was quickly found that the s.e. of all three unknowns decreased as the flattening was increased and that the sums of the squares of the residuals went through a minimum when the flattening was ~ 0.012 – 0.014 . The results are

Table 1 Computer fits

Adopted flattening	Equatorial radius (km)	Correction to RA (s)	Correction to dec. (")
0.0052	$3,458.4 \pm 3.6$	$+0.00173 \pm 21$	$+0.0145 \pm 135$
0.0097	$3,461.6 \pm 2.3$	$+0.00185 \pm 13$	$+0.0115 \pm 87$
0.0135	$3,464.4 \pm 1.9$	$+0.00195 \pm 11$	$+0.0089 \pm 71$

given in Table 1. The residuals of the observations were very good—the greatest residual in the radius being 3.9 km, which corresponds to a timing residual of only 0.18 s.

The values for the equatorial radius have not been corrected for refraction. To obtain the true distance from the centre of Mars the scale height should be added. This is probably ~ 10 km, but will doubtless be determined more accurately from the individual light curves at a later date: it should then be possible to improve the solution by applying the scale height to each observation individually.

Table 2 Flattening and equatorial radii at various levels

Level	Flattening	Equatorial radius (km)	Method
Surface	0.0057 ± 0.002	$3,394 \pm 4.5$	Spacecraft radio occultation ⁴
?	0.0091 ± 0.002	$3,402 \pm 16$	Visual micrometer observations ⁵
0.5 light	0.013	$3,474 \pm 3^*$	Photoelectric occultation

*Includes an estimated error for the scale height.

Table 2 shows the flattening and equatorial radii at various levels. For comparison purposes the radius derived from the photoelectric observations has been increased by an assumed scale height of 10 km.

It may eventually be possible to express scale height as a function of latitude. Since the observations at each phase range over only 25° in latitude, however, it is unlikely that there will be a marked change in the flattening.

The cause of this observed flattening may partly arise from a diurnal variation in density—the disappearance phase occurred on the unilluminated limb while the re-appearance phase occurred on the illuminated limb. No such effect, was, however, detected when Jupiter occulted a star in 1971, the flattening at 0.5 light level being very close to the dynamical flattening⁶.

In conclusion, it would seem from this analysis that the 0.5 light level has an equatorial radius of $3,464 \pm 2$ km (+ scale height) and a flattening in the range 0.012 to 0.014.

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Hydrodynamics and geodynamics in the Phlegraean fields area of Italy

WE show here, from an analysis of geological observations in the Phlegraean Fields, that the high correlation among slow ground motions, seismicity and short and long period tidal components (including the Chandler wobble) observed at Pozzuoli (Naples) suggests that the sea, throughout intercrustal

hydrodynamic correlation, may be an important geodynamic agent.

The intense Phlegraean–Vesuvian volcanic event occurred in Campania (southern Italy) in the area lying between Monte Massico, the margins of the pre-Apennine arc, the Sorrento peninsula and the Gulfs of Naples and Pozzuoli¹. The Phlegraean Fields are the volcanic district W of Naples (Fig. 1a), some of whose craters form part of the city's urban system. The Phlegraean volcanic activity, as opposed to that of Vesuvius which is famous for its lava manifestations, is mainly characterised by huge explosions of the phreatic type, with ejection of predominantly ash products. A thick buildup of which is followed by intense diagenesis with the formation of the classic Phlegraean lithoid tuffs. The characteristics of these tuffs are their considerable cohesion and very high porosity (40% at the surface); consequently their rheological behaviour can be compared with that of a sponge dipped in water².

The morphology of the Phlegraean Fields is characterised by shallow craters sometimes affected by marine erosion, by many thermal springs both on land and under water, some feeding the

Fig. 1 Phlegraean Fields volcanic basin. *a*, First-order residual gravity anomalies. Also shown: survey points; morphological and structural borders of the caldera; the west–east and north–south trends of the profiles of Fig. 1b and Fig. 4 respectively. *b*, Profiles of the maximum uplift (—▲—▲—▲) and residual gravity anomalies (—), with the geological cross section according to the west–east trend.

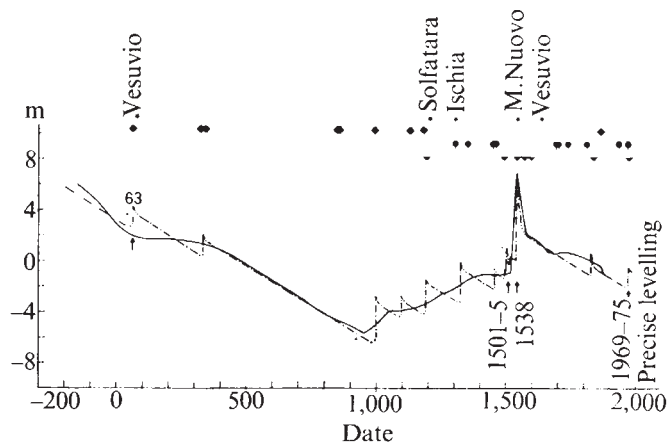
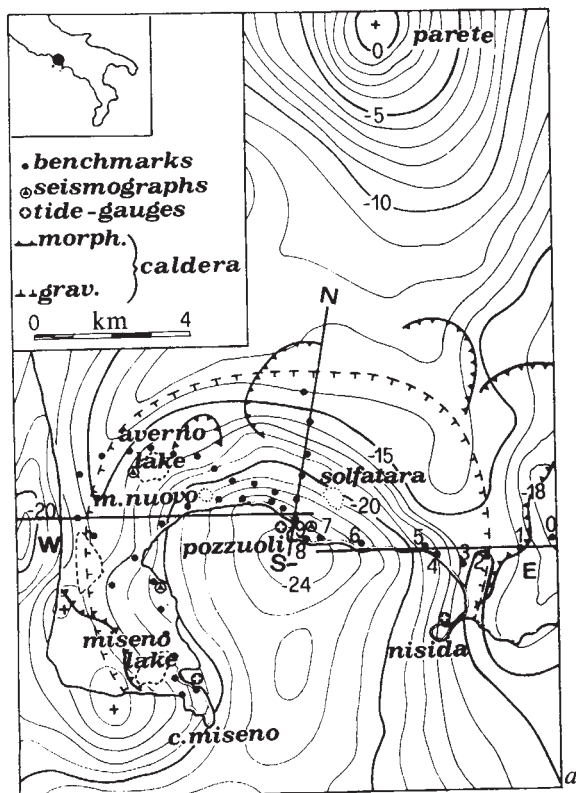
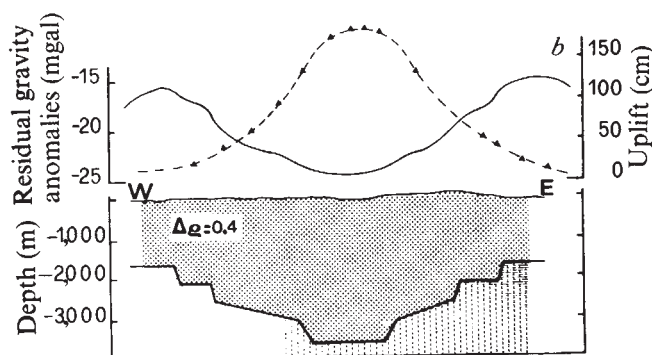


Fig. 2 Trend of Phlegraean bradyseism over the centuries. (—) Parascandola's trend; (---) represents the modified curve. ↑ gives date of historical uplift; *, a major eruption in Campania; ◆, a major earthquake in Campania; ●, a major earthquake in Campania, also causing damage in Naples; ▼, an earthquake in Pozzuoli.

spas that were already famous in Roman times, and by solfatara fields. The dynamics of the Phlegraean Fields is characterised by a vertical movement of the crust to which has been attributed the Greek term 'bradyseism'. The latter brought about one of the longest series of continuous up-and-down movements. In fact, the trend of this local vertical movement, that has continued for many centuries at a time, has been reconstructed from the study of the clear traces left by the lithodomes on the surface of the columns of the Roman *Mercatum*, called Serapeo, built in the second century BC at Pozzuoli.

Figure 2 shows the diagram drawn, as an unbroken line, by Parascandola³, which is also based on the data collected by Niccolini⁴ in the middle of the nineteenth century. It is particularly interesting to note that in 1538 there was a ground uplift of ~ 7 m in only 48 h when the eruption of Monte Nuovo took place, and that the maximum uplift was recorded in Pozzuoli at ~ 3 km from the point of eruption.

In the study of the origin of Phlegraean bradyseism² the lowering phenomenon is interpreted as being a result of compaction through overloading of the light pyroclastic products filling the Phlegraean basin down to a depth of 3.5 km (gravimetric model shown in Fig. 1b).

The uplift, on the other hand, is interpreted as being the result of the rheological behaviour of the porous fluid-saturated media affected by the flow variation in these same fluids as a consequence of both a variation in the amount of heat carried up from below (increase in the convective movements) and the seismoeruptive effects of the phreatic explosion.

Moreover, as it is well known that earthquakes are also accompanied by ground uplift events, which, although much smaller than those recorded for the Phlegraean area, have similar characteristics, Parascandola's diagram has been suitably modified as shown in Fig. 2 with the dash and dot line.

The proposal to modify the trend of the historical data was made on the assumption that when the local and regional eruptions and earthquakes take place there may also be discontinuity, with a rapid ground uplift and a successive return to subsidence conditions. Only the eruptions of Vesuvius of the years AD 79 and 1631 have been considered, as they began new eruptive cycles after a few centuries of inaction.

The hypothesis that the Phlegraean uplift movements take place over short periods of time is confirmed by the recent inversion of bradyseism noted in early February 1970, whose development is shown in Fig. 2.

The recent event, which probably began in 1969 and continued for four years, has enabled the study of some of the characteristic manifestations.

The inversion was seen to be accompanied by a continual increase in steam flow from the fumaroles and a widening of

Solfatara di Pozzuoli mud pool. The increase and decrease of these reflected the ground uplift trend. No variation in the temperature of these same manifestations was ever observed.

The data we have analysed were gathered by several companies and organisations⁵ and covered the following: ground movement, microseismicity and sea movement.

In the case of the ground uplift that took place from March 1970 to February 1973 the Campanian Public Works Office carried out precision level measurements on several tens of benchmarks throughout the Phlegraean zone. The heights had been checked various times between 1903 and 1953. Along the Naples (Fuorigrotta)–Pozzuoli coastline the measurements made on benchmarks numbers 0–9 were made on average at monthly intervals. The resulting uplift data were found to have both area (Fig. 1b) and time dependencies (Fig. 3). Figure 3a shows that the ground movement trends with respect to the maximum height reached in the individual benchmarks are in close agreement with one another.

The maximum uplift was reached in July 1972 (Fig. 3) and the average trend for benchmark 7 (where the absolute maximum was reached) can be expressed by a typical diffusion equation

$$q = q_0 + m \exp[-\lambda_1 (t_0 + t)] - n \exp[-\lambda_2 (t_0 + t)]$$

where q is the height reached, m , n , λ_1 and λ_2 are constants

Fig. 3 Time dependence of the ground uplift in the Phlegraean Fields. *a*, Variation (with respect to March 9, 1970) in the height of the benchmarks assuming for each one the height reached on July 6, 1972 (referred to as q_m in the figure) to be 1.0. Benchmark 0 is regarded as being stable. This shows the average value for each series of measurements and the interval (I) containing the random distribution of the single values from the 9 benchmarks. For benchmark 0, q_m was 0 (cm); for 1, 3.00 cm; for 2, 8.5; for 3, 17.36; for 4, 28.75; for 5, 35.89; for 6, 69.51; for 7, 70.37; for 8, 68.46; for 9, 64.60. *b*, High accuracy levelling values for benchmark 7. Ordinates (origin at the level of May 19, 1969) are the lower set on the left and go on near the curve. *c*, Best fit curve (see text); *d*, *e*, *f*, trends obtained from tidal data given in Corrado and Palumbo⁸. Ordinates are on the right (the origin is not at the same level as the *b* and *c* curves).

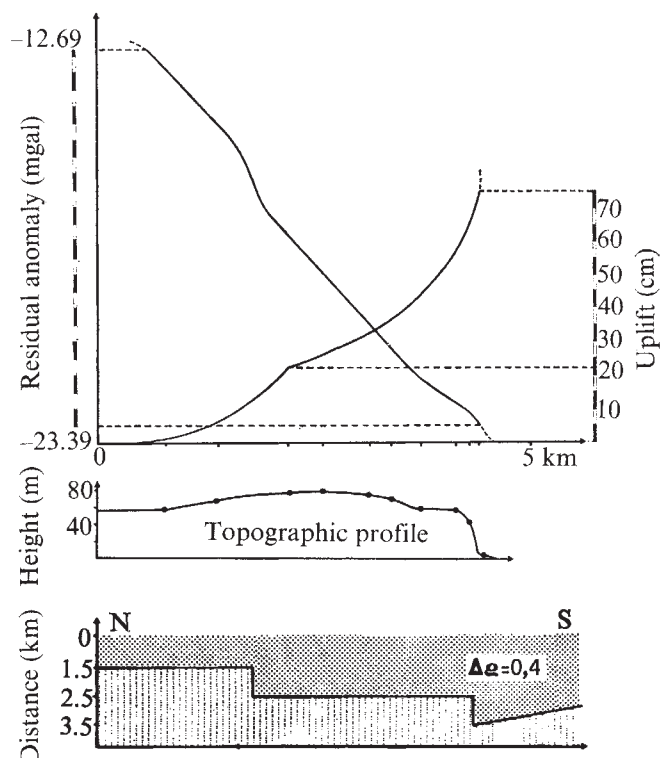
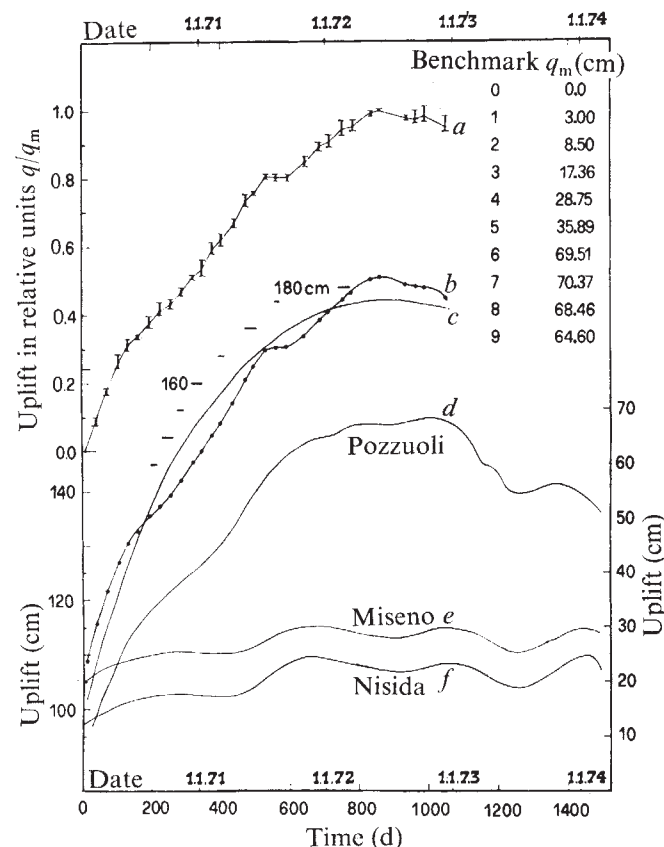


Fig. 4 Trend of uplift from 1962 to 1970 and (n-1) the order gravimetric residual anomalies; topographic profile and geological cross-section along the N–S profile indicated in Fig. 1a (after Montagna⁷).

chosen to give the best fit to the experimental data; t_0 is when the bradyseism inversion began ($\sim$ May 19, 1969; $t_0 = 285$ d) when $q = -23.77$ cm, compared with the height measured in 1953 by I.G.M. This is in agreement with the dependence of uplift on the variation of the flow of fluid within porous layers (Fig. 3c).

The uplift profile (Fig. 1b) shows an increase from the borders of the caldera inwards, that is, from almost negligible values to 156 cm compared with the height in 1953 (180 cm compared with May 1969 when the inversion of the phenomenon probably began). The uplift values can be connected with different thicknesses of the pyroclastic layer (Fig. 1b).

During the recent uplift, the altimetric survey made by the Italian Geological Survey in 1962 (ref. 6) along the N–S profile of Fig. 1a was repeated (April 1970) and a more detailed gravimetric survey⁷ was also carried out. The two-dimensional model, drawn from the first-order residual anomalies, compared with the height variations, shows a clear correlation between the type of uplift and the thickness of the pyroclastic products covering the limestone substratum. Figure 4 reveals the correspondence between the uplift discontinuities and the position of the faults⁷.

These observations are in agreement with the data on the ground movement obtained⁸ by comparing the average sea level far away (the Naples and Pozzuoli tide-gauging stations).

The correlation coefficient between the data in Fig. 3b and those in Fig. 3d was found, by Pearson's formula of the product moment, to be 0.994.

The curves shown in Figs 3 and 5c reveal that the uplift in the different stations in the Phlegraean Fields between 1970 and 1975 has a trend which is the combination of different periodical components.

Three seismic stations have been in operation⁵ since March 1970 to study the seismicity in the Phlegraean Fields. At the Pozzuoli station (Vescovado: Fig. 1a), where the majority of the seisms have been recorded, 5,752 seismic events were noted up to February 1974. The foci were essentially concentrated within a depth of 3.5 km and had a random distribution.

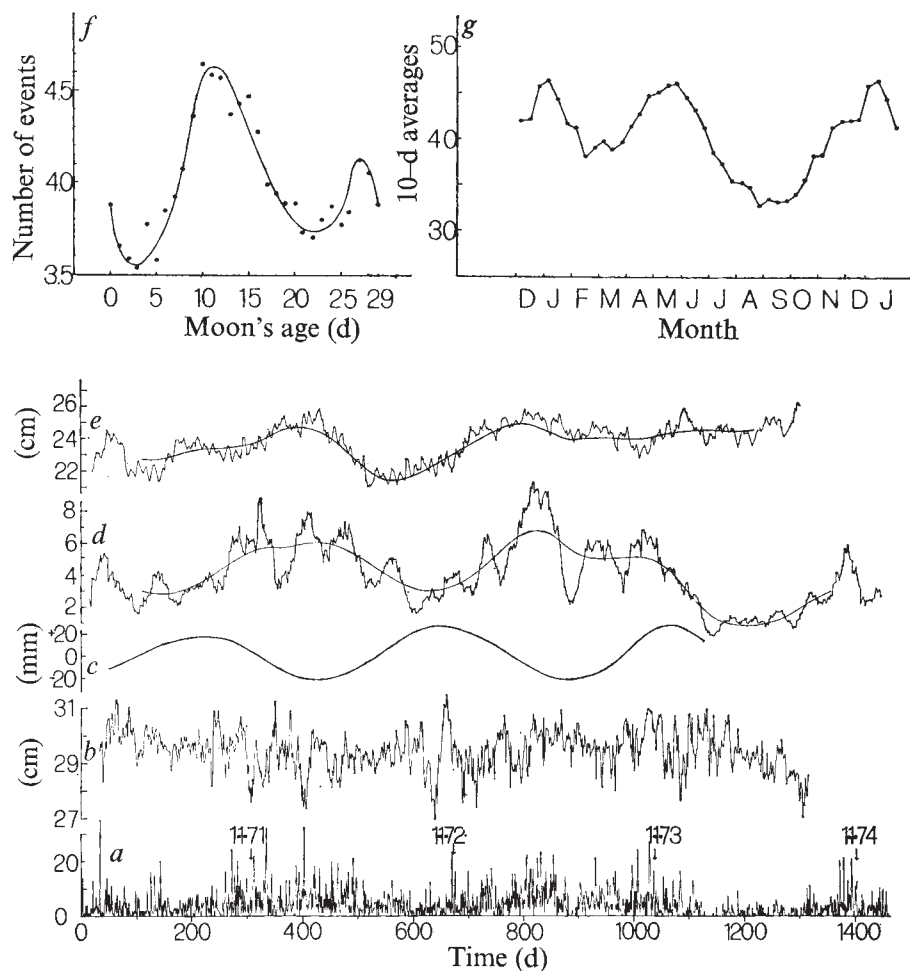


Fig. 5 Trend of seismicity, vertical ground movement and sea movement at Pozzuoli. *a*, Number of daily seismic events recorded at Vescovado station; *b*, Average daily sea level; *c*, Ground uplift calculated by subtracting the average trend values from those of the curve in Fig. 3*d*; *d*, average daily number of seismic events for 29 consecutive days along with the relative average trend; *e*, analogous values and average curve for the tide amplitude; *f*, average daily seisms in function of the Moon's age for the period 1970–74; *g*, average 10-d seisms for the same period.

Only about ten of these were felt by the local population, having reached an intensity of III–IV Mercalli degrees.

Also the daily trend of the seisms is clearly periodical (Fig. 5*a* and 5*d*): the long period trend is accompanied by several short period trends, in particular the fortnightly (Fig. 5*f*) and six-monthly (Fig. 5*g*).

The average daily sea level (Fig. 5*b*) and the sea-level variation over 6 h (an index of its mean velocity), obtained by subtracting the low tide level from the previous high tide level (Fig. 5*e*), refer to Pozzuoli station (Capitaneria di Porto). The latter curve also shows, besides the smaller period trends, a clear long period one.

The correlation coefficient of the average curve drawn from Fig. 5*d* and *e* is 0.81, also calculated from Pearson's formula: the 99% confidence limits are 0.69 and 0.89.

This suggests that there is a direct connection between the sea motion, considered as a 'tidal pumping', and the micro-fracturing of the skeleton of the highly porous and permeable pyroclastic layer.

The trend of the uplift curve (Fig. 5*c*), obtained at Pozzuoli by subtracting the average uplift values from the values of the curve in Fig. 3*d*, is analogous to those of Fig. 5*d* (daily seismic events) and 5*e* (tide amplitude) and the correlation coefficient, as calculated once again from Pearson's formula, between values in Fig. 5*c* and *d* is 0.73: the 99% confidence limits are 0.84 and 0.57.

In spite of the short interval considered (1,400 d) the fact that the three phenomena have a common period of 400–450 d brings to mind the Chandler wobble. To confirm this connection the analysis of tidal and geodynamic (earthquake and vertical ground movement) data should cover a period of at least 7 yr.

In conclusion, it would seem that the phenomena accompanying the Phlegraean bradyseism give support to the interpretation of its genesis² based on the alteration of intercrustal

hydrodynamics, and in particular they suggest that the sea has an influence on local geodynamics.

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Ophiolite emplacement on to continental margins

RECENT work on west Newfoundland ophiolites supports Elliott's¹ contention that the mechanics of emplacement of oceanic crust and upper mantle on to continental margins ('obduction') is no different from the emplacement of thrust sheets in any foreland thrust belt. Evidence to this effect comes from the presence of minor structures found along the thrust fault surfaces separating the various structural slices, as well as from other related minor structures—hitherto unrecorded in ophiolite suites—that have been found within the thrust sheets themselves. These

reveal a very specific kind of information about the mechanics of ophiolite transportation and, without reference to the forces causing emplacement, they imply that many of the processes involved in the motion of ophiolite thrust sheets are equivalent to those typical of foreland thrust belts—due consideration being given to basic differences in the material properties of the rocks involved.

As far as the basal metamorphic aureole associated with the western Newfoundland ophiolites is concerned, it is clear that it predates their transportation on to the continental margin. Direct evidence to this effect is found in the Bay of Islands complex² where the low greenschist grade thrust faults that are responsible for ophiolite transportation obviously truncate the higher grade aureole, placing it and the adjacent ophiolite stratigraphy structurally over and above the unmetamorphosed oceanic sedimentary rocks. In other places this low greenschist grade structural base may follow the pre-existing metamorphic aureole³, using it as a suitable discontinuity, analogous to a bedding plane thrust in a foreland thrust belt; although this would appear to obscure the history and chronology of the two events (that is, an earlier one that produced the higher grade metamorphic aureole, from a later one that emplaced the ophiolites on to the continental margin along greenschist grade thrust faults), a distinction can be made on the basis of minor structures observed within the thrust sheets themselves. These take the form of narrow ductile deformation zones (DDZ, see Fig. 1) formed during the motion of the ophiolite sheet; locally these clearly truncate and retrograde to greenschist facies the granulites and amphibolites of the basal aureole, as well as parts of the overlying ophiolite stratigraphy. Such relations express, rather explicitly, the lack of significance of the basal metamorphic aureole in terms of the immediate processes inherent to the orogenic belt in which the ophiolites have been emplaced. In terms of its pre-emplacement significance, one wonders if the high temperature aureole might be explained as the product of dynamic conditions induced during seafloor spreading, by the lateral flowage of oceanic crust and upper mantle in the vicinity of an ocean ridge? Or, could it be the vestige of a subduction zone fabric?

With respect to greenschist grade emplacement mechanics, the presence of narrow ductile deformation zones throughout a large volume of an ophiolite thrust sheet shows that there is a fundamental difference in the internal energy dissipation⁴ within an ophiolite thrust sheet, compared to that of a foreland thrust sheet involving only unmetamorphosed cover rocks. In the latter, a large proportion of energy is absorbed by folding, generally taking advantage of the facilitating slip planes provided by well developed bedding. In contrast, in the massive or crudely layered crystalline rocks which constitute most of an ophiolite sheet, deformation is much less homogeneous, taking place along narrow, often distantly spaced, ductile deformation zones. Their number and intensity varies according to position within the ophiolite thrust sheet: although no attempt has yet been made to quantitatively evaluate this, clearly they are much more widely spaced and weakly developed throughout the bulk volume of the thrust sheet than near its base. In the proximity of the fault plane (within 30 m) there is a seemingly exponential increase in the number of DDZ; at the fault, the deformation zones run together, with small ones feeding larger ones—in effect, emptying their displacement cumulatively into large shear zones which subsequently empty theirs into the main thrust fault at the base of the ophiolite slice. Similar ductile deformation zones have been described by Ramsay and Graham⁵, and Mitra (unpublished) in deformed crystalline basement and intrusive rocks of several orogenic belts. Their significance here, in terms of ophiolites, lies in their recognition for the first

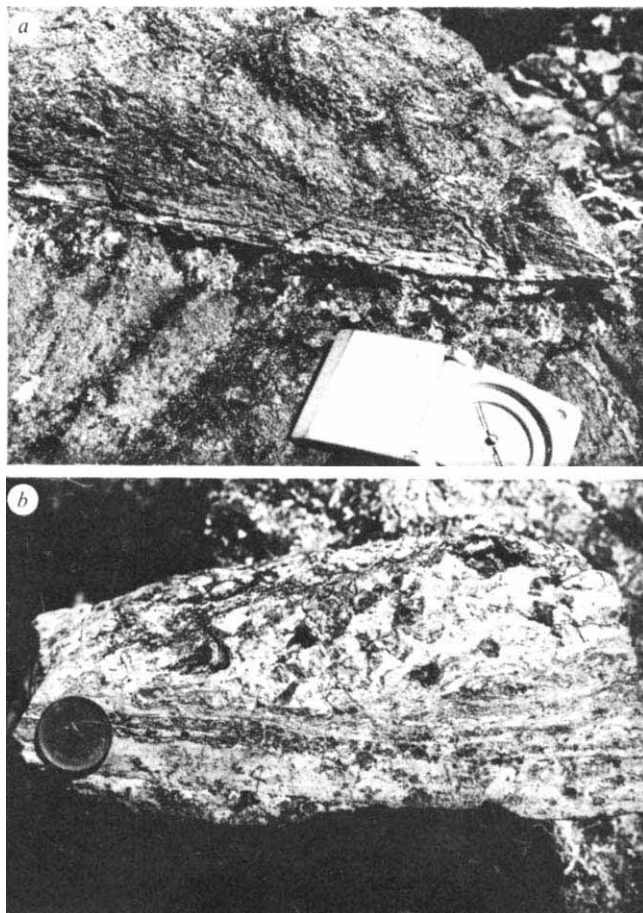


Fig. 1 *a*, Asymmetric ductile deformation zone cutting across the gabbro cumulates, east side of Blow Me Down Mountain, Bay of Islands complex. *b*, Close-up of a sample from the same deformation zone, showing the spectacular grain-size reduction of the original mineralogy (coarse pyroxenes in feldspar matrix) to a narrow, greenschist-grade mylonite.

time as diagnostic minor features related to the transportation and emplacement of oceanic crust and mantle on to a continental margin. Although in terms of internal energy dissipation they record a fundamental difference between ophiolite and foreland thrusts, this is only important in consideration of the material properties inherent to the specific thrust sheet.

Information on the physical mechanisms operative at the base of a moving ophiolite thrust has been recorded by a variety of minor structures along the actual fault surface. These include: (1) fibrous accretion steps (micro-lite fibres where ultramafics occur in the hanging wall), and (2) striations, gouge, and polished surfaces, similar to those features described by Elliott. These are indicative of pressure solution slip and frictional sliding processes operating at the base of a moving ophiolite sheet, as in the case of a foreland thrust⁴.

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Water movement in porous media towards an ice front

In frozen soils water movement towards an ice front can occur, leading to the formation of ice lenses, frost heaving and damage to roads and other structures. During the past decades many attempts have been made to explain the transport phenomena involved, fundamental contributions being made by Everett¹ and Takagi², among others. We present here experimental results on the influence of pressure on ice growth; they accord with thermodynamic theory.

At our laboratory Vignes and Dijkema³ studied the flow of water towards an ice front through a very narrow slit (50 nm × 1 mm × 50 mm), all walls consisting of quartz. They found that under isothermal conditions the flow occurred at a rate proportional to the degree of subcooling. This result was confirmed at our laboratory by De Loos (internal report) who studied flow of water towards an ice front through a glass cylinder of 1 mm² cross section, containing 10⁴ parallel cylindrical capillary channels of 1 µm diameter.

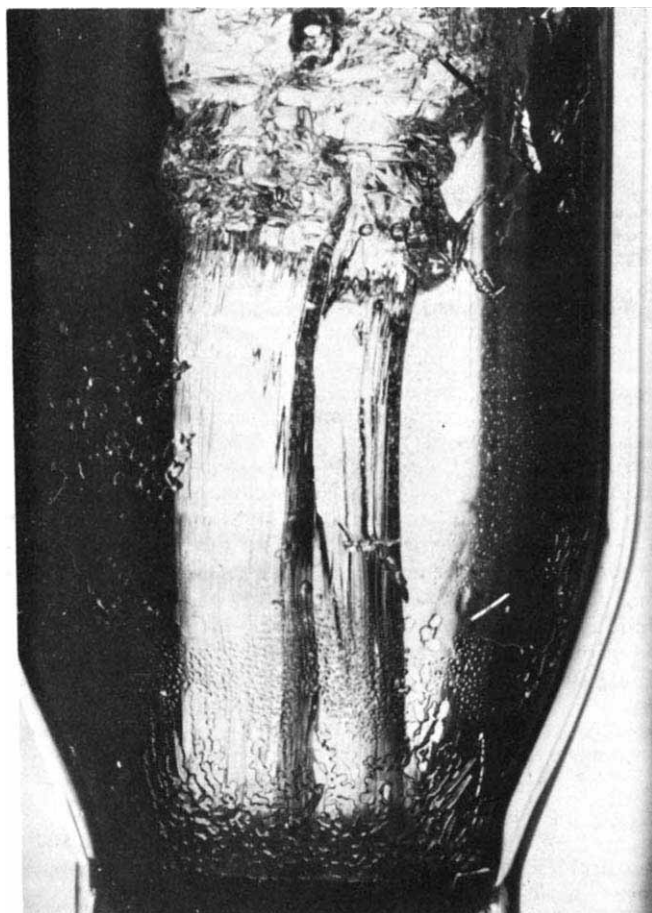


Fig. 1 Ice column (diameter ≈ 7 mm) formed during 15 d after starting ice growth. $T = -0.045^\circ\text{C}$, $p_i = p_w = 101$ kPa.

Vignes and Dijkema³ explained their experimental results by suggesting that the seat of the phenomenon is to be found in the thin layer of unfrozen adsorbed water between the ice and the quartz wall. The pressure distribution in this layer is thought to be anisotropic, leading to heaving of the ice by the disjoining pressure on the one hand, and water flow by suction in a direction parallel to the wall on the other hand. This was

elaborated on by Vignes⁴ in a theoretical study, treating the thin water layer as a micropolar fluid.

In subsequent experiments we extended this work by investigating the influence of pressure on the rate of water movement. Ice and super-cooled water were separated by a horizontal glass filter, all walls consisting of glass as well. The ice was situated on top of the filter. The pressure of the water and the ice could be controlled independently.

Under isothermal conditions, and at atmospheric pressure on both ice and water, water flow towards the ice was again observed. Figure 1 shows an ice column formed during 15 d of stationary conditions.

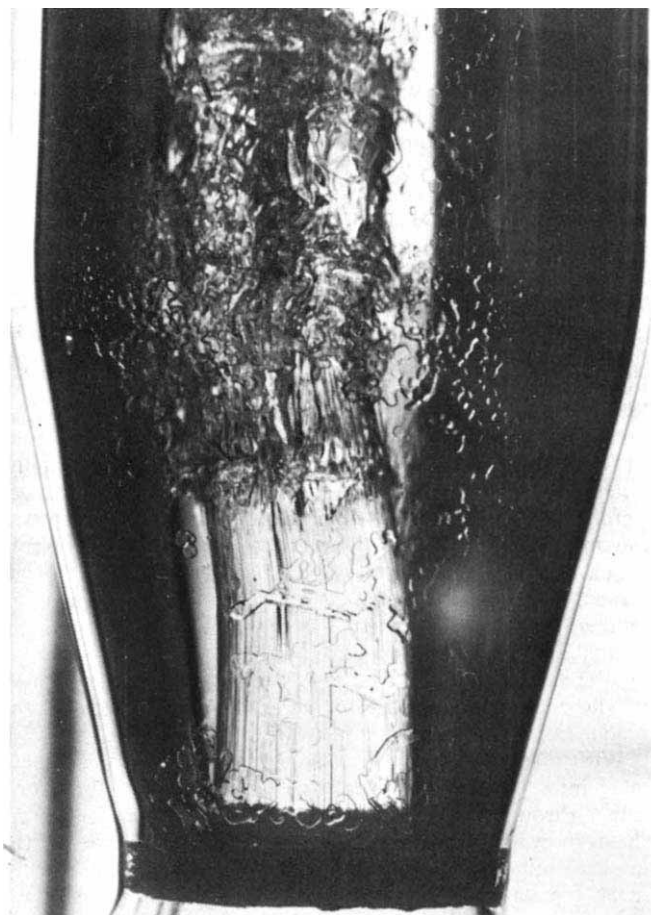


Fig. 2 Photograph taken at condition of no water flow. $T = -0.05^\circ\text{C}$, $p_i - p_w = 63$ kPa, $p_i = 101$ kPa.

Decreasing the water pressure at constant temperature, while the pressure on the ice remained at 1 atm, led to a decrease of the rate of water movement and eventually to a reversal of its direction, accompanied by melting of the ice. This is illustrated in Figs 2 and 3; it can be clearly seen that the ice column melts at the bottom.

The pressure difference between ice and water at equilibrium (no water flow) is found to obey closely the equation

$$p_i - p_w = -h(T - T_0)/v_w T_0$$

where the subscripts i and w refer to ice and water, p is pressure, T temperature, T_0 equilibrium temperature at $p_i = p_w = 1$ atm, h specific enthalpy of fusion, and v specific volume. This equation can easily be found by equating the differentials of the chemical potentials of ice and water and the subsequent



Fig. 3 Photograph taken 1 d after that of Fig. 2. $T = -0.05^\circ\text{C}$, $p_i - p_w = 76\text{ kPa}$, $p_i = 101\text{ kPa}$.

integration for constant h and v_w . The temperature range investigated was -0.01°C to -0.06°C .

A full account of this work will be published elsewhere.

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Inaccuracy in rainfall measurement

RAINGAUGES are intended to give accurate measurements of precipitation. There is, however, a growing interest in the chemical composition of rainwater, and in the effects of chemicals in rain on plants and soil. It is also sometimes realised that the water collected in raingauges may be contaminated by dry deposition on the collecting funnels, and also by chemicals derived from the materials used in constructing these funnels and the collecting bottles beneath them.

For this latter reason, raingauges may be constructed of high density polythene, or of polypropylene, materials found not to contaminate seriously the water they collect. The collecting funnels of these raingauges may be cleaned at various intervals by different workers, in an attempt to reduce surface contamination. Unless the cleaning is done in a standard way at a standard time, the chemical analyses of different samples may be difficult to compare. It is fairly

common practice to clean funnels once a week, when samples are being taken. This may mean that in dry weather all surface contamination is lost, while if it rains just before the surface is cleaned much of the soluble material on the funnel's surface will be washed into the collecting bottle.

I now find that cleaning the polypropylene funnels may also affect the volume of the samples of rain collected. Funnels which have been exposed for several months, and which have been washed only by the falling rain, allow all the precipitation falling on them to run quickly down into the collecting bottles. The same happens with dew. These 'dirty' funnels measure the rainfall accurately.

When, however, the funnels are cleaned by daily wiping with paper tissue dampened with distilled water, they behave quite differently. Their surface is seen to be covered with beads of water immediately after rain, or in the early morning when there has been a heavy dew fall. This water does not run down into the collecting bottle, and is rapidly evaporated and lost when the weather is dry.

The amount of water lost may not be insignificant. The polypropylene funnels I use have a diameter of 20 cm. This means that they collect 31.4 ml of water for each mm of rainfall. Shortly after an occasion where there was a rainfall of 0.8 mm, the 'dirty' funnels were almost completely dry, having transferred 25 ml of water to their collecting bottles. The 'clean' funnels were covered with drops of water. These were mopped up with paper tissues, which were then weighed. The water on the funnels was found to amount to between 5 and 6 ml. With this slight amount of rain the shortfall in collection amounted to some 20%. In heavy rain the loss is not, proportionately, so great, but it is still significant.

This behaviour of the rain gauge surface will not only affect the volume of water collected; it will also affect the way in which the surface deposition is dissolved. A heavy dew or a slight shower which runs off the 'dirty' funnel will remove much soluble deposition, while on a 'clean' funnel, if the dew or rain is not collected, any salts dissolved will be redeposited when the water evaporates, and they will not appear in the sample which is collected and analysed.

It thus seems that, if we are interested only in accurate measurements of the amount of rainfall, we should not clean the surfaces of our collecting funnels. If we are interested in rainwater chemistry, we should plan our work knowing that the results may be affected by the treatment given to the collecting surfaces.

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Gas velocities inside a burning cigarette

To understand more fully the factors that contribute to the formation and mass transfer of products within a burning cigarette, and to specify the type of reactions occurring, it is necessary to know the velocity of gases inside the combustion coal. In this study, velocities have been calculated from pressure and temperature distributions inside the burning cigarette. Gas velocities as high as 400 cm s^{-1} occur within the coal during a puff. Thus the residence times of gases in the coal can be much lower than 1 ms. This time is not long enough to allow molecular reactions of primary products in the gas phase to contribute to the final products.

The direction of the flow of gases inside the cigarette can be obtained from the internal pressure distribution, since the gas flow is perpendicular to the pressure contours and is towards

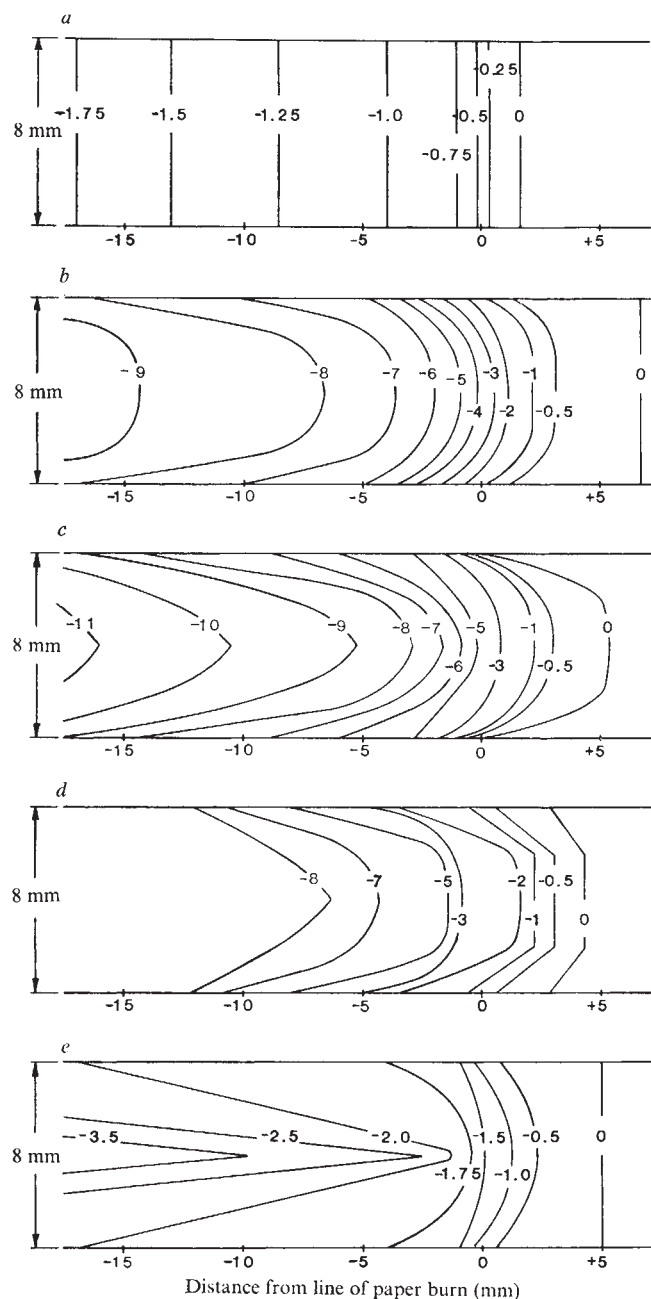


Fig. 1 Pressure (cm water) distribution in the cigarette at various times after the start of a 2-s, 35 cm³ puff (with a square pressure-time puff profile). Axial positions in the unburnt tobacco rod are quoted as negative distances from the burn line, while positions in the coal and ash are quoted as positive distances. *a*, 0.1 s; *b*, 0.5 s; *c*, 1.0 s; *d*, 2.0 s; *e*, 2.1 s.

lower pressures. Furthermore, inside a cigarette the volumetric flow of gases (V , cm³ s⁻¹) and pressure differential (ΔP , cm water) across a given length (L , cm) obey Darcy's Law¹

$$V = -\frac{1}{\varepsilon} A \frac{\Delta P}{L} \quad (1)$$

where A is the area through which the gas flows (cm²), ε is the impedance of the tobacco bed at room temperature (cm water s cm⁻²), and 1 cm water = 98 N m⁻².

The impedance of a body is directly proportional to the viscosity (η , poise) of the fluid flowing through the body². A previous study has confirmed that the structure of the tobacco bed does not change significantly with temperature¹, so that ε is a function of η only. Consequently, if the flowing gas is at

temperature T , from equation (1) the component of velocity u_x (cm s⁻¹) in any direction is given by

$$u_x = -\frac{1}{\varepsilon} \frac{\eta_0}{\eta_T} \frac{\partial P}{\partial x} \quad (2)$$

where η_0 and η_T are the viscosities of the gas at room temperature and T , respectively.

Thus, since the temperature distribution of the gas phase inside a burning cigarette is known³, the gas velocities inside the cigarette can be obtained from the pressure distribution using equation (2), assuming that the viscosity of the gases inside the cigarette vary with temperature as does air. This assumption is not critical, since the major gas phase components inside the cigarette (nitrogen, water, carbon monoxide, carbon dioxide and oxygen) all have viscosities whose variation with temperature closely parallels that of air^{4,5}.

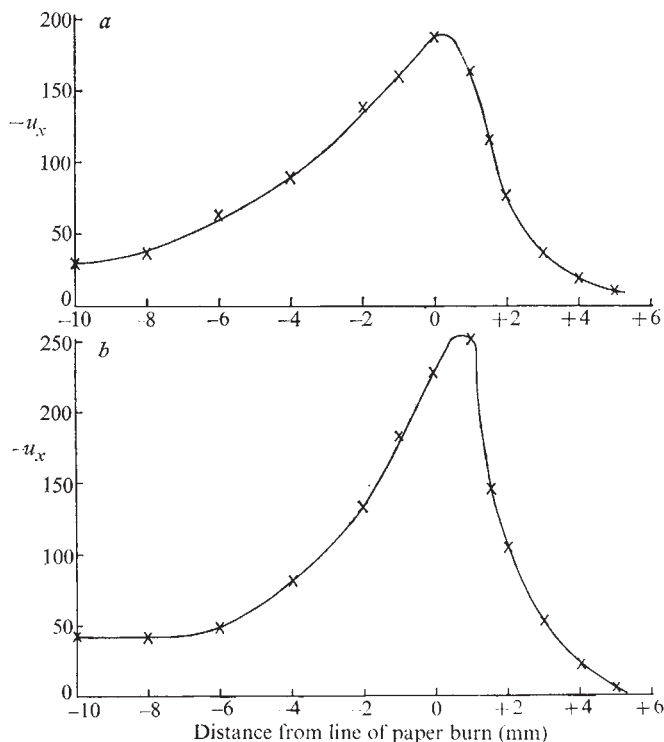
The pressure distribution inside the cigarette during the third puff of the smoking cycle has been determined. A quartz glass tube, 0.80 mm internal diameter, 1.12 mm outer diameter, and 35 mm long, was inserted radially into the cigarette, while its other end was connected to an Ether type UPI pressure transducer. The output from the pressure transducer was monitored on a CAMAC data logger. The resolution of a pressure measurement was ± 0.01 cm water. The transducer-probe system gave 100% response to a pressure change from ± 10 cm water (relative to atmospheric pressure) to atmospheric pressure in under 0.1 s.

The cigarettes used, and the experimental and computational techniques, were similar to those described previously for determining the temperature distributions³.

The pressure contour distributions during the puff are shown in Fig. 1. The mean pressures have 95% confidence limits of up to 25% of their value, due to slight variations in the cigarettes used. Consequently, the quoted pressure distributions are very much mean distributions, and the distribution in an individual cigarette could be significantly different.

The mean pressure contours 0.1 s after the start of the puff are all perpendicular to the cigarette axis (Fig. 1*a*). Thus, the

Fig. 2 Gas velocity (u_x , cm s⁻¹) along the central axis of the cigarette at *a*, 0.5 s; and *b*, 1.0 s after the start of a 2-s puff. The cigarettes had an impedance of 0.042 cm water s cm⁻².



gas flow through the cigarette is entirely parallel to the cigarette axis. Furthermore, the flow only extends to a distance of 1.5 mm in front of the line of paper burn.

At 0.5 s after the start of the puff, the pressure within the cigarette decreases further, air leakage through the paper causes convex shaped contours behind the coal, measurable negative pressures extend for up to 6.6 mm in front of the paper burn line, and the pressure contours in the coal are convex (Fig. 1b). Consequently, at this time in the puff, a proportion of the air enters the coal in an approximately radial direction, just in front of the paper burn line. As the puff progresses (Fig. 1), the pressure contours in the vicinity of the paper burn line tend to become more nearly parallel to the

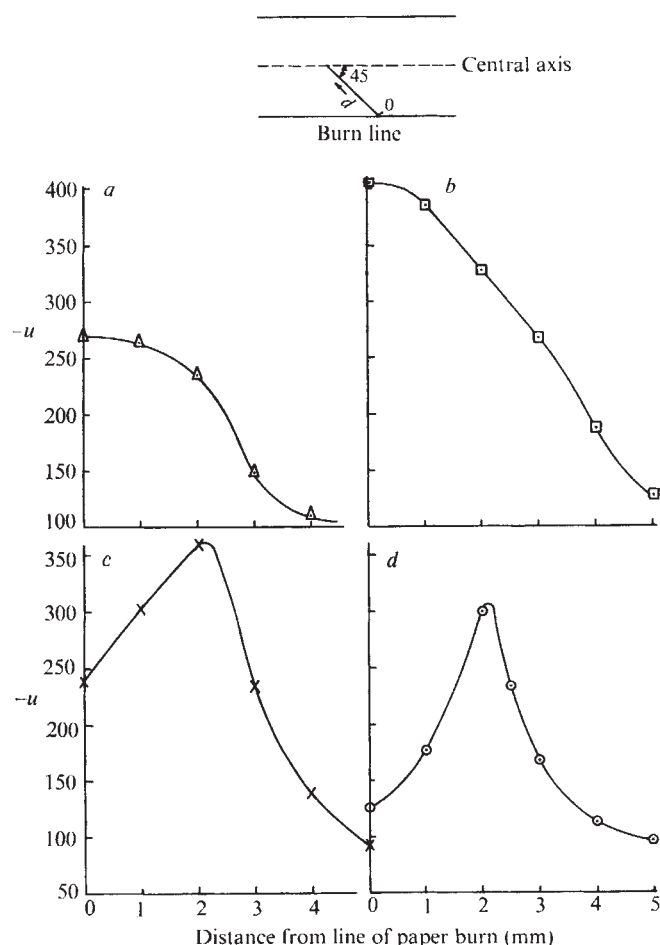


Fig. 3 Gas velocity (u , cm s⁻¹), d mm from the paper burn line, along a line at 45° to the central axis, at various times during a 2-s puff. a, 0.5 s; b, 1.0 s; c, 1.5 s; d, 2.0 s.

cigarette axis, indicating that the air influx in this region becomes more nearly radial. Thus, during a puff, air initially enters the coal in a radial direction in the vicinity of the paper burn line, and its line of flow gradually becomes axial behind the coal. Consequently, it is largely the peripheral region of the coal that burns during a puff, as has previously been observed by direct studies⁶.

The pressure distribution 0.1 s after the end of the puff is shown in Fig. 1e. The pressures inside the cigarette have increased considerably (become less negative) in the ensuing 0.1 s, particularly at the peripheral regions of the cigarette behind the paper burn line. Thus, when the puff ends, air must enter the cigarette predominantly through the paper. Within 1 s of the puff ending, no mean negative pressures are observed within the cigarette.

The gas velocities along the central axis of the cigarette during a puff, calculated using equation (2) and data from Fig. 1 and ref. 3, are shown in Fig. 2. At distances greater than 20 mm behind the line of paper burn, where the flowing gases are close to room temperature, the linear gas velocity along the central axis is 30–42 cm s⁻¹ at all times during the puff. The greatest linear velocities along the central axis (160–250 cm s⁻¹) are obtained just in front of the line of paper burn (Fig. 2).

Gas velocities in any direction of interest can be calculated. Inspection of Fig. 1 suggests that a major route for gas intake into the coal throughout the puff is at an angle of 45° to the central axis, and intersecting the paper at the burn line. Calculated velocities along this line are given in Fig. 3, and it is seen that 1.0 s after the start of the puff, gas velocities in excess of 400 cm s⁻¹ are obtained. Along this line, 1.0 s after the start of a puff, the gas temperature falls from 520 °C at the burn line to 100 °C 1.4 mm away³. Consequently, the incoming gases have been heated to 520 °C, and then cooled to 100 °C, in 350 μ s. Molecular reactions of intermediate volatile products are almost certainly too slow to occur in the residence times available in the hot coal. This means that further reaction of primary decomposition and combustion products in the gas phase must occur by radical reactions and possibly by ionic reactions as well.

It is interesting to note in Figs 1 and 2 that measurable pressures and central axis gas velocities only extend to distances of about 5 mm in front of the paper burn line during the puff. Hot gases (temperatures greater than 100 °C), however, extend to distances of 11–13 mm in front of the paper burn line³. Consequently, diffusion processes, and not forced flow processes, are the major mechanism by which the mass transport of gases occurs in the coal at distances greater than about 6 mm from the paper burn line.

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Dependence of induced mutation on spontaneous mutation in *Tribolium castaneum*

IN plants the effect of physical and chemical mutagens has been found to depend on the frequency of spontaneous mutation^{1,2}, but no such information is available from animal material. I have therefore investigated the relationship between spontaneous and X-ray-induced somatic mutation in the flour beetle *Tribolium castaneum*. This relationship was found to be positive and significant when the genotype was manipulated by artificial selection for speed of development.

The criterion used consisted of the wing developmental abnormalities that are under genetic control (21 genes have been identified, all with a recessive mode of inheritance³) and are subject to environmental modification^{3–5}. The data are the same as those for the lines selected for speed of

development⁴. Beetles were irradiated as 1-d-old pupae with 10 kR and the induced somatic mutations were observed in the adults which emerged. Spontaneous mutations were observed in non-irradiated samples. The experimental conditions were maintained at 33 °C and 70% relative humidity.

Figure 1 shows that the rate of induced mutation was dependent on that of spontaneous mutation among wild, slow and fast lines of beetles. The degree of determination of R^2 is very high (0.994), indicating that variation in the induced mutations could be explained by variation in their spontaneous rates.

Because the rate of mutation for wing abnormalities is under genetic control and is subject to natural and artificial selection^{5,6}, both types of mutation could be an expression of common underlying processes in the enzymatic system acting on the regulatory mechanism which controls DNA synthesis and repair⁷. The slow line is expected to have the lowest rate of both spontaneous and induced mutation, for slowing of development gives more time for repair to take place⁴. The wild type, the best fit genotype, has higher mutation rates than both the selected strains. These results confirm that the optimal mutation rate within wild strains is favoured by natural selection⁸ and is independent of the rate of lethal mutation, as a high mutation rate is expected to have deleterious effects⁹. On the other hand, the relationship between fitness (% adult emergence) and mutation rate (% wing abnormalities) for eight wild strains of *T. castaneum* was negative¹⁰ ($r = -0.82$, $P < 0.05$).

A line selected for intermediate development values (stabilising selection) showed a higher degree of spontaneous mutation and a slower degree of induced mutation than did

Fig. 1 Relationship between X-ray-induced mutations and spontaneous mutations of three lines selected for seven generations for speed of development and their control line. Means $\pm$ s.e. are based on four replications. Correlation and regression coefficients are based on the angular values of the means of the fast, slow and control lines (connected by the straight line). Analysis of variance on such values indicated highly significant differences between lines and between the two types of mutations, but not between replications nor sex or for the interaction components. Numbers are the total number observed. $r = 0.997$; $P = 0.05$; regression coefficient = 3.275, s.e. = 0.242.

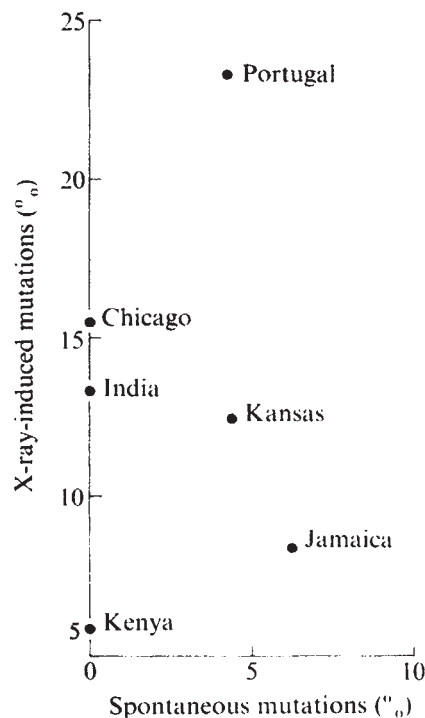
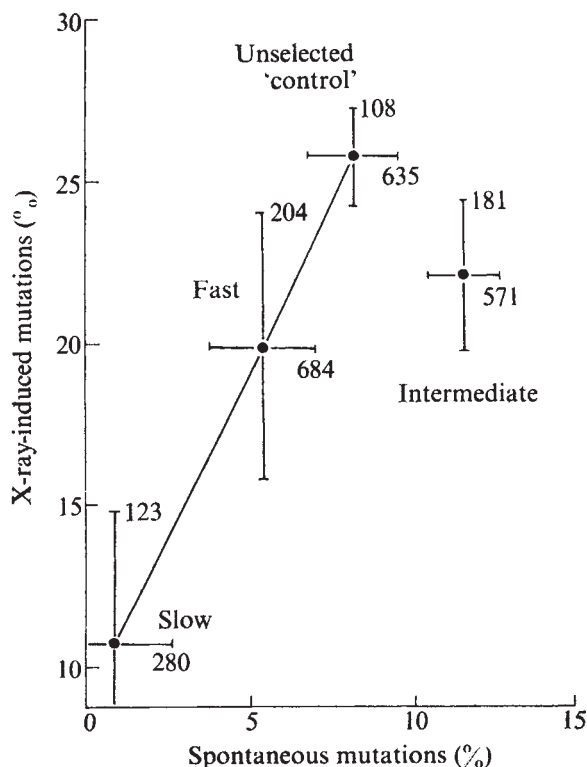


Fig. 2 Scatter diagram for the relationship between the two types of mutations of six wild populations of *T. castaneum*.

the control line (Fig. 1). This indicates that stabilising selection is efficient in producing genotypes that could stand sudden environmental changes (X ray), while these genotypes are less stable in normal environmental conditions. In other words, X rays have more effect on the wild strain (due to a certain degree of homozygosity) than the intermediate line (due to developmental homeostasis); while in stable environments spontaneous mutation is more pronounced in the intermediate line (due to induction of new recessive mutants) than the wild strain (due to greater genetic homeostasis). The different degrees of genetic and developmental homeostasis among different wild populations with different genetic backgrounds will undoubtedly affect the relationship between spontaneous and induced mutations observed among these populations (Fig. 2).

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Artificial deciliation causes loss of calcium-dependent responses in *Paramecium*

WHEN a paramecium collides with an obstacle, it reverses the direction of the effective stroke of its cilia, swims backwards for a while, and, with one or more repetitions of this 'ciliary reversal' response, avoids the obstacle¹. When the medium is disturbed² or when the posterior part of its body is stimulated mechanically, it swims faster than usual and

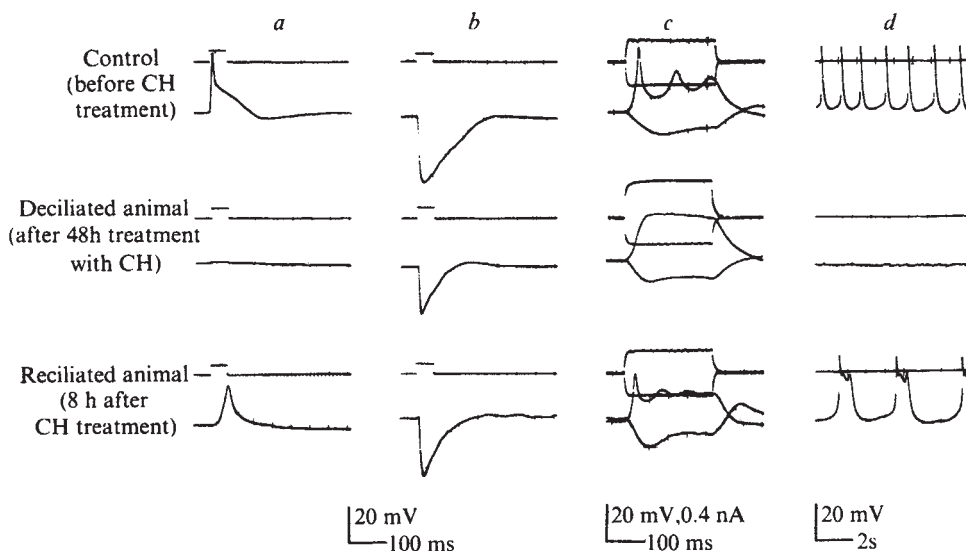


Fig. 1 *P. caudatum*, reared in hay infusion and washed in the adapting solution, which consisted of 2 mM CaCl_2 , 1 mM KCl and 1 mM Tris-HCl, pH 7.4, was deciliated with CH, which was added to the adapting solution at 5 mM. The animals were kept for about 48 h in this solution at 20 °C. *a*, *b*, and *c*, Membrane potentials were recorded in the adapting solution at 20 ± 2 °C; *d*, recordings were made in a solution of 2 mM BaCl_2 , 1 mM CaCl_2 and 1 mM Tris-HCl, pH 7.4, 20 ± 2 °C. For recording, a glass capillary microelectrode, filled with 0.3 M KCl and with a resistance of 200–400 M Ω was used. Specimens that showed gradual depolarisation of the resting membrane potential were discarded. Successful recordings were made from about 120 specimens. *a*, Responses to mechanical stimuli given to the anterior end of the animal by means of a glass stylus with a rounded tip mounted on a piezoelectric phonograph cartridge. In each record the upper trace indicates the zero potential level as well as the electric pulse used to drive the stylus. *b*, Responses to mechanical stimuli given to the posterior end. The methods of stimulation and recording are the same as in *a*. *c*, Responses to direct current pulses applied through an intracellular electrode. The upper traces in each record indicate both the zero potential level and the applied current. In the case of the deciliated animal, the current was increased to exclude the possibility of change in the threshold of the regenerative response. *d*, Spontaneous membrane activity in Ba^{2+} - Ca^{2+} mixture. The upper trace in each record shows the zero potential level.

escapes from the stimulus mainly by increasing the frequency of ciliary beat. The current view of the underlying mechanisms can be summarised as follows^{3,4}. A mechanical stimulus to the anterior end of the animal deforms the cell membrane of this region, increasing the permeability to Ca^{2+} and causing a depolarisation which further activates the Ca^{2+} -channels in the membrane. Thus, the depolarisation develops regeneratively towards the equilibrium potential of Ca^{2+} . The Ca^{2+} which has entered the cell acts on some mechanism controlling the direction of ciliary beat to bring about ciliary reversal. On the other hand, a stimulus to the posterior end activates the K^{+} -channels in the membrane, causing hyperpolarisation and an increase in ciliary beat frequency. Although the animal behaves as a mechanoreceptor cell in the early stages of these events, it is not clear which part of the cell transduces the mechanical stimulus to the activation of Ca^{2+} - or K^{+} -channels. We report here the effect of removal of cilia on the electrophysiological responses of paramecium to mechanical as well as some other stimuli.

Paramecium caudatum was deciliated by treatment with chloral hydrate (CH) according to the method of Kuźnicki⁵. The methods of stimulation and recording were similar to those of Naitoh and Eckert⁶. Typical results are shown in Fig. 1.

Figure 1*a* and *b* shows membrane responses to mechanical stimuli, given by a fine glass stylus to the anterior and posterior ends of the animal, respectively. Untreated paramecia responded with depolarisation to anterior stimulation and with hyperpolarisation to posterior stimulation. When the animals were incubated in medium containing 5 mM CH, they lost the ability to swim within 24 h and sank to the bottom of the incubating glassware. Under a light microscope, the cilia appeared unimpaired in both number and individual length at this stage, but their movement was uncoordinated. Electrophysiological responses to mechanical stimulation were not significantly different from those of the untreated animal. As incubation continued, the cilia began to drop off, and in about 48 h all were lost except for

those in the oral groove. The deciliated paramecia retained the hyperpolarising response to posterior stimulation, but did not show a depolarising response to anterior stimulation. A stronger stimulus (obtained by pushing the stylus from a shorter distance) caused hyperpolarisation even if given to the anterior end. This eliminates the possibility of a general decrease in responsiveness. The resting membrane potential was not affected by CH. If a deciliated paramecium was transferred to medium containing no CH, cilia regenerated completely in 6–8 h. The reciliated paramecia responded with depolarisation to anterior stimulation, although both the amplitude and the rate of increase of the response were smaller than normal for some hours after the apparent completion of reciliation.

The finding that the depolarising responses disappear and reappear in parallel with the loss and the regeneration of cilia can be interpreted most readily if it is assumed that the Ca^{2+} -channels are localised in the ciliary membrane, whereas the K^{+} -channels are not. Although the possibility cannot be ruled out that CH inhibits Ca^{2+} -channels in the extraciliary membrane independently of the deciliating effect, it seems unlikely that the time course of such inhibitory effects coincides with that of the deciliating effect. The interpretation that the Ca^{2+} -channels are lost by deciliation is supported by two further experiments.

Figure 1*c* shows responses to current pulses applied through a second intracellular microelectrode. Fluctuations in potential (peaks) superimposed on the passive depolarisation are seen in the untreated animal. These have been ascribed to the regenerative activation of Ca^{2+} -channels⁷. This response also disappeared with deciliation and reappeared with reciliation. Similar results have recently been reported by Dunlap and Eskert^{8,9} who found that deciliation by vigorous agitation of *P. caudatum* incubated in CH eliminates the 'calcium response' to a depolarising current pulse, while regrowth of the cilia restores the response. They also interpret the results as indicating the localisation of Ca^{2+} -channels in the ciliary membrane.

Figure 1*d* shows spontaneous periodic depolarisations

observed in a solution containing Ba^{2+} and Ca^{2+} . This response is believed to depend on Ba^{2+} moving into the cell through the Ca^{2+} -channels¹⁰. The Ba^{2+} -response also disappeared and reappeared in parallel with deciliation and reciliation, although the frequency and the form of potential changes were altered somewhat in the reciliated animal.

Electron microscopic studies on paramecia have shown that binding sites for divalent cations are localised in the basal region of each cilium¹¹⁻¹³. Plattner suggested that the granular patches of these divalent cation-binding sites have a role in the regulation of ciliary movement by binding and releasing Ca^{2+} . It is interesting that with CH treatment the cilium is lost just proximal to these structures¹⁴. Although there is no evidence that these Ca^{2+} -binding sites are identical with the Ca^{2+} -channels, it is tempting to relate our results to these morphological findings.

During the preparation of our manuscript we learnt that Dr K. Dunlap, University of California, Los Angeles (personal communication), also observed, in *Paramecium caudatum*, loss of the regenerative component of membrane response by CH deciliation and its recovery with reciliation.

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Two populations of heterozygote erythrocytes in moderate hypoxanthine guanine phosphoribosyltransferase deficiency

THE X-linked condition hypoxanthine guanine phosphoribosyltransferase (HGPRT) deficiency may be either gross or moderate in degree and is invariably associated with overproduction of urate in the hemizygote^{1,2}. In addition, however, the gross deficiency state (Lesch-Nyhan syndrome)³ also manifests with mental deficiency, choreoathetosis, spasticity and self-mutilation. Fibroblasts from heterozygotes for the gross deficiency state display mosaicism, some having normal HGPRT activity and others the reduced HGPRT activity characteristic of the hemizygote⁴. However, HGPRT activity in erythrocyte lysates from such heterozygotes is normal. Heterozygotes for the moderate HGPRT deficiency state, on the other hand, have HGPRT activity in erythrocyte lysates ranging between 20% of normal and completely normal values⁵. To elucidate this, we have developed an autoradiographic procedure to show HGPRT activity in individual erythrocytes. In families with moderate HGPRT deficiency, in whom erythrocytes of the hemizygote were not significantly labelled by this procedure, heterozygotes showed two erythrocyte populations, one deficient in HGPRT and one containing HGPRT activity. On the other hand, in families whose HGPRT deficiency was gross and manifested as the Lesch-Nyhan syndrome, heterozygotes showed only the normal pattern of labelling. Thus, mosaicism was present in erythrocytes from heterozygotes for moderate, but not for gross, HGPRT deficiency. The former

finding supports the Lyon hypothesis⁶ whereas, the latter finding suggests that either the X chromosome carrying the mutant HGPRT gene is preferentially inactivated, or the X chromosome is randomly inactivated with later selection against the erythrocyte precursors containing the mutant HGPRT enzyme.

Normal nucleated cells incorporate tritiated purine bases into nucleic acids, permitting autoradiographic demonstration of the presence of HGPRT activity⁷. Erythrocytes also contain considerable amounts of HGPRT and, although they are capable of accumulating significant quantities of labelled mononucleotides, they do not synthesise nucleic acids. Earlier attempts to demonstrate HGPRT activity in erythrocytes by autoradiography failed because the soluble mononucleotides, unlike nucleic acids, are lost immediately the selective permeability of the cell membrane is destroyed by fixing or drying the cells. Thus, the demonstration of HGPRT activity in erythrocytes by autoradiography using the conversion of tritiated hypoxanthine to nucleotide required fixation of the labelled inosinic acid *in situ*. We have done this by precipitation of the mononucleotides by lanthanum, as developed by Bakay⁸, simultaneously with fixation of the red cells by osmium tetroxide. After autoradiography, silver grains were counted over 200 cells and the distribution of the grain counts per cell was plotted. The distribution of counts in normal subjects showed that at least 90% of erythrocytes had more than five grains per cell; in Lesch-Nyhan subjects, no erythrocytes had more than this value. The grain counts per cell were therefore combined into three groups (Fig. 1). To validate the method, the distribution of grain counts was studied in artificial mixtures of erythrocytes from a normal subject and one with the Lesch-Nyhan syndrome. The results enabled the proportions of the two types of cell to be confirmed (Fig. 1) and thereby justified the search for two populations of erythrocytes in heterozygotes.

Members of one family with the Lesch-Nyhan syndrome and three families with less severe HGPRT deficiency were studied by this technique (Fig. 1). The HGPRT activity in cell-free lysates of erythrocytes and fibroblasts from hemizygotes and heterozygotes for each of these families is shown in Table 1. In family W (Lesch-Nyhan syndrome)⁹, erythrocytes from two obligate heterozygotes, both of whom showed normal activity in an erythrocyte lysate, showed a uniform pattern of labelling indistinguishable from normal. In hemizygotes from families L and C¹⁰, erythrocytes were indistinguishable from those of the Lesch-Nyhan syndrome. The heterozygote in family L, whose HGPRT activity in an erythrocyte lysate (Table 1) indicated that, if there were two populations, 90% of her erythrocytes would contain mutant HGPRT enzyme*, showed 86% of

Table 1 Hypoxanthine phosphoribosyltransferase activity in cell lysates (nmol per mg protein per h)

Family		Erythrocytes	Fibroblasts
W (Lesch-Nyhan)	Hemizygote	< 0.01	< 1
	Heterozygote 1	89	70
	Heterozygote 2	85	88
L	Hemizygote	< 0.05	7
	Heterozygote	10	103
C	Hemizygote	< 0.05	27
	Heterozygote	44	46
B	Hemizygote	13	30
	Heterozygote	18	60
Normal $\pm$ s.d.		101 $\pm$ 12	157 $\pm$ 32

*Percentage of erythrocytes containing mutant HGPRT enzyme =

$$\frac{A_N - A_H}{A_N - A_m} \times 100$$

where A_N is HGPRT activity in normal erythrocyte lysate, A_m is activity of mutant HGPRT enzyme in erythrocyte lysate from hemizygote, and A_H is HGPRT activity in erythrocyte lysate from heterozygote.

erythrocytes with less than five grains per cell. In the heterozygote from family C, the expected percentage of HGPRT-deficient erythrocytes was 56% and a value of 70% was observed. These results were taken to confirm the presence of two populations of erythrocytes in heterozygotes for the moderate HGPRT deficiency syndrome.

The fourth family, B, had the least severe HGPRT deficiency (Table 1) and the least severe clinical manifestations¹⁰. However, the pattern of grain counts over the erythrocytes of the hemizygote was close to normal, although there were more cells with less than five grains per cell than were seen normally. The autoradiographic procedure was therefore modified by reducing the duration of incubation and extending the exposure time. This modification did not significantly alter the pattern of labelling in normal erythrocytes, but it resulted in 86% of the erythrocytes from the hemizygote showing less than five grains per cell. Erythrocytes from the heterozygote of this family also showed a similar proportion of HGPRT-deficient cells, a finding expected from the HGPRT activity of her erythrocyte lysate. This technique has therefore demonstrated the expected

proportion of normal and HGPRT-deficient erythrocytes in heterozygotes from families with moderate HGPRT deficiency. It would seem that in the functionally less severe HGPRT mutations, random inactivation of the X-chromosome occurs and two types of erythropoietic clones result, one of which produces normal erythrocytes and another which produces erythrocytes containing mutant HGPRT enzyme.

In the severe HGPRT deficiency of the Lesch-Nyhan syndrome, on the other hand, although the technique could identify the reduced HGPRT activity of the mutant enzyme, no HGPRT-deficient erythrocytes were detected in two heterozygotes whose heterozygosity had been established by both the production of affected sons and autoradiography of fibroblasts. Although two populations of fibroblasts are found in heterozygotes for the Lesch-Nyhan syndrome, all previous studies have demonstrated normal HGPRT activity in erythrocyte lysates, and several other studies have suggested that the X chromosome of all of their erythrocyte precursors was normal¹¹⁻¹³. The present technique provides direct confirmation of this. Either inactivation of the X chromosome is not random in these cells

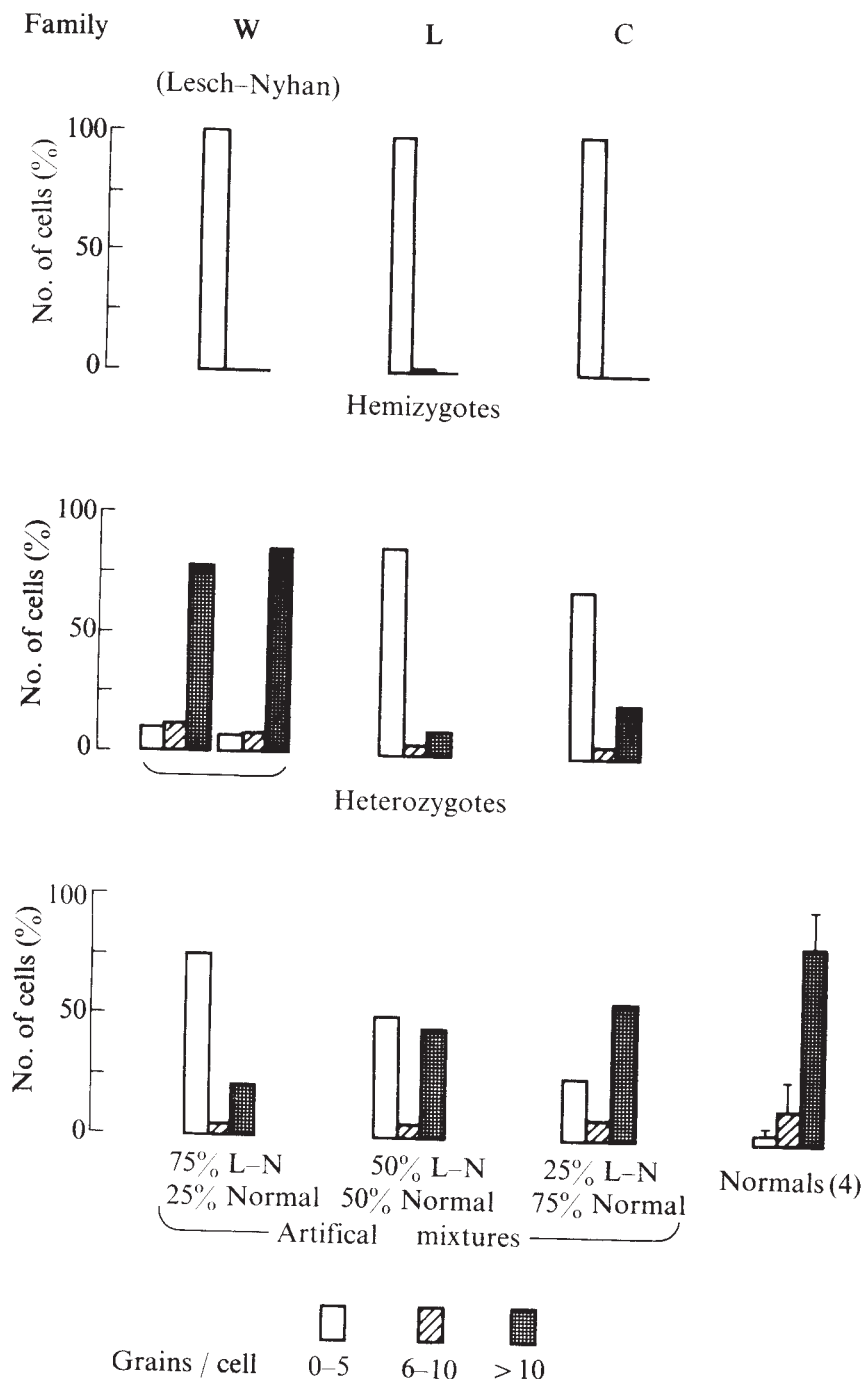


Fig. 1 Comparison of grain counts per cell in autoradiographs of erythrocytes from normal subjects, from hemizygotes and heterozygotes from three families with HGPRT deficiency and in artificial mixtures of normal and HGPRT-deficient erythrocytes. Erythrocytes were incubated with labelled precursor in a special incubation chamber constructed by cementing a glass ring to a cover slip with Araldite. The incubation mixture contained 0.05 M sodium phosphate (pH 7.4), 0.075 M sodium chloride, 5 mM glucose and 0.05 mM ³H-hypoxanthine [¹ Ci mmol⁻¹ (Amersham) 99% pure on electrophoresis]. Aliquots (200 μ l) of the incubation mixture were pipetted into the incubation chamber, and 5 μ l of a 1:100 saline dilution of heparinised and washed packed erythrocytes was added and mixed by swirling. An acid-washed glass microscope slide was then placed over the incubation chamber, and the whole slide and chamber were then inverted and incubated in an humidified atmosphere at 37 °C for 30 min. During the incubation, the cells settled and became firmly attached to the slide; the glass ring and coverslip prevented spreading of the cell suspension. The wide separation of cells on the slide also prevented any possible cell-to-cell transfer of labelled material. After incubation, the chamber was removed and the slide immediately washed thoroughly to remove all traces of labelled hypoxanthine, care being taken to cover the cells at all times with a layer of saline. After washing, the slides were flooded with a fixative consisting of 1 volume of 0.1% osmium tetroxide and 9 volumes of 0.12 M neutral lanthanum chloride (0.12 M lanthanum chloride neutralised with lanthanum oxide and filtered). After fixation for 15 min, the slides were washed repeatedly with distilled water and allowed to dry. The dry slides were immersed in absolute methanol for 1 h, washed with methanol and dried. The slides were then coated with photographic emulsion (Kodak NTB 2) and exposed for 3 d. After development, the slides were stained with 0.5% aqueous eosin for 10 min, washed with water, destained in a solution containing 50% methanol and 50% citrate phosphate buffer (4 volumes 0.1 M citric acid + 6 volumes 0.2 M disodium hydrogen phosphate) for about 15 s and immediately washed with absolute methanol and dried. The autoradiographs were permanently mounted before grain counting. Control slides using normal erythrocytes were examined with each batch of cells studied.

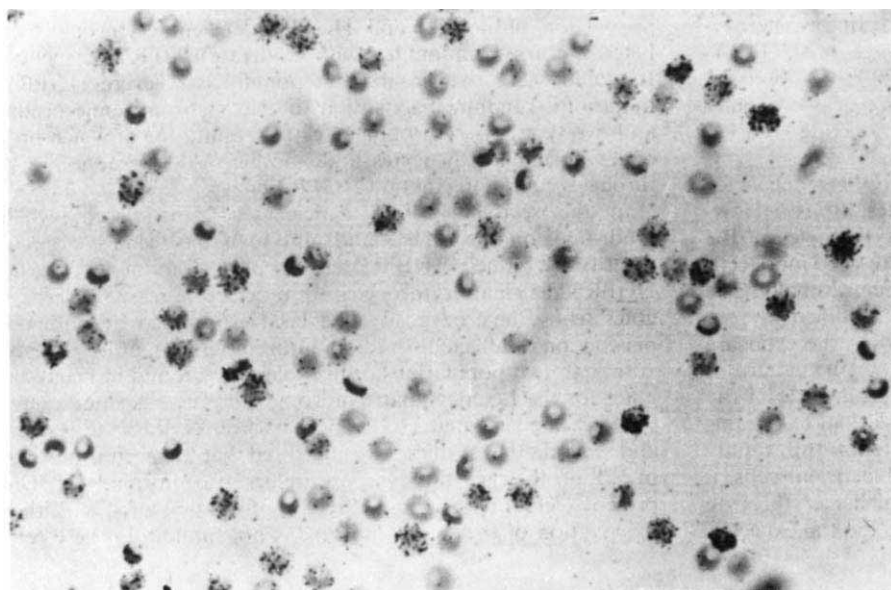


Fig. 2 Autoradiograph of erythrocytes from the heterozygote of family C showing populations of labelled and unlabelled cells.

or there is subsequent selection against haemopoietic precursor cells containing mutant HGPRT enzyme. Support for such selection against HGPRT-deficient stem cells *in vivo* in heterozygotes for the Lesch-Nyhan syndrome, has been provided by studies of the culture characteristics of their bone marrow cells¹⁴. Our findings further suggest that, whereas there exists a wide spectrum of severity of HGPRT deficiency, there is a critical intracellular level of HGPRT activity which, in the hemizygote, leads to the Lesch-Nyhan syndrome and which, in the heterozygote, renders affected erythrocyte precursors at a marked functional disadvantage in comparison with the normal.

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Glucocorticoid receptors in murine embryonic facial mesenchyme cells

IN various experimental animals, clefting of the developing secondary palate can be induced by the administration of glucocorticoids to pregnant animals¹⁻⁴. Various inbred strains of mice exhibit different degrees of susceptibility to glucocorticoid-induced cleft palate⁵⁻⁸. Specifically, A/J mice

treated with glucocorticoids between days 11 and 14 of gestation (day of detection of vaginal plug designated day 0 of pregnancy) produce 100% of their offspring with cleft palate, whereas C57BL/6J (C57) mice similarly treated produce offspring with only 20-25% cleft palate. Although it has been demonstrated that glucocorticoids can elicit divergent physiological responses in various differentiated cell types through a class of specific cytoplasmic and nuclear receptor proteins⁹⁻¹¹, the exact molecular mechanism(s) by which glucocorticoids function as teratogens as well as the aetiology of the strain-specific difference in the response to glucocorticoids are unknown. We present here evidence that mouse facial mesenchyme cells obtained either directly or in primary culture possess specific glucocorticoid-receptor proteins of high affinity and limited capacity for dexamethasone. Furthermore, we report that A/J facial mesenchyme cells possess approximately double the amount of receptor proteins than C57 mesenchyme cells.

Embryos were obtained from day 14 pregnant A/J or C57 mice (purchased timed pregnant from Jackson Laboratories and obtained on day 11 of pregnancy). Embryos were dissected from the uteri in phosphate-buffered saline (PBS). Placentae and other extraembryonic membranes were removed, and the embryos were washed in PBS. Secondary palates or maxillary processes were dissected from the embryos and pooled. Minced tissues were dissociated with 0.25% trypsin (Difco) containing 0.1% EDTA (Fisher Scientific) for 10 min at 37 °C on a shaking water bath. After dissociation, NCTC-109 medium (Microbiological Associates) containing 10% foetal calf serum (Gibco), streptomycin (100 µg ml⁻¹, Gibco) and penicillin (100 units ml⁻¹, Gibco) was added at 4 °C to inhibit the trypsin. Cells were dispersed, washed and seeded into 75-cm² tissue culture flasks (Falcon) at a density of 5.4 × 10⁴ cells per cm². Cells were grown in NCTC-109 medium containing 10% foetal calf serum and antibiotics. Cultures of mesenchyme cells obtained from either the secondary palate or maxillary processes appeared to have a fibroblastic morphology.

Cell suspensions were incubated for 40 min at 37 °C with ³H-dexamethasone to measure glucocorticoid receptor activity since fluorinated glucocorticoids in contrast to the naturally occurring corticosteroids do not bind to serum transcortin (CBG)¹⁴, a component of foetal calf serum. Under these conditions, steroid uptake was maximum for both A/J and C57 mesenchyme cells. Figure 1 illustrates one of several experiments in which the specific binding of ³H-dexamethasone to whole cell suspensions of A/J or C57

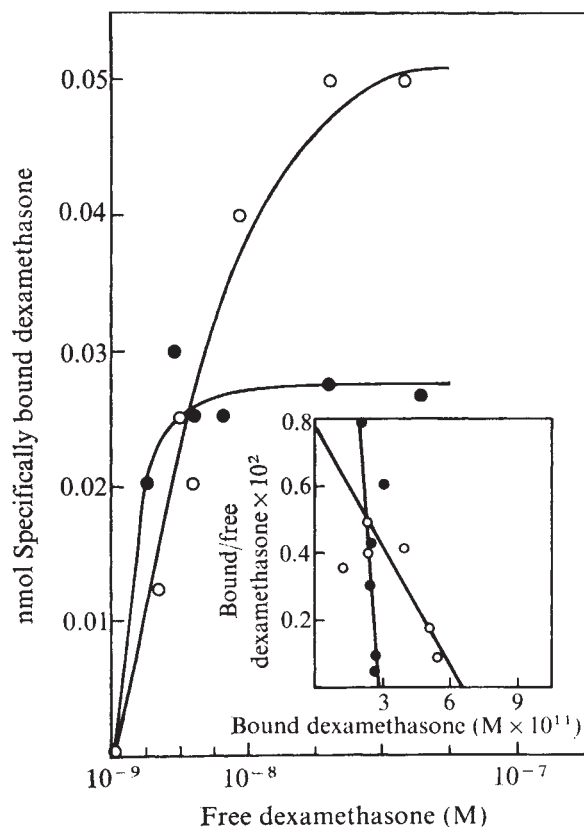


Fig. 1 At confluency (approximately 4–5 d after seeding), cells (1.5×10^6 per cm^2 at confluency) were trypsinised (0.1% trypsin), centrifuged and washed in Dulbecco's modified PBS at 4°C (Ca^{2+} and Mg^{2+} free, 0.01% EDTA, 0.02% dextran). Glucocorticoid-receptor activity of both cytoplasmic and nuclear origin was measured in whole cell suspensions using 1, (2)- ^3H -(N)-dexamethasone (specific activity 28 Ci mmol^{-1} ; Amersham/Searle) as a probe according to the method of Baxter *et al.*¹² as modified by Sibley and Tomkins¹³. Approximately 3×10^6 cells obtained from A/J (○) or C57BL/6J (●) maxillae were incubated in 0.5 ml warm (37°C) serum-free NCTC-109 medium containing 1–60 nM ^3H -dexamethasone in the absence or presence of an excess of unlabelled dexamethasone (10^{-5} M) in parallel assays to correct for nonspecific binding¹². Cell suspensions were incubated at 37°C for 40 min on a shaking water bath and vortexed periodically. At the end of the incubation, aliquots were removed from each assay to calculate the total concentration of ^3H -dexamethasone, and the suspensions were chilled to 4°C . All subsequent procedures were carried out at 4°C . The cells were centrifuged at 800g for 5 min. The pellets were washed with 5.0 ml of PBS, recentrifuged, and resuspended in 0.5 ml of distilled water. The pellets were then freeze-thawed three times in an ethanol-dry ice bath. Aliquots of the lysates were either taken in duplicate and dissolved in 10 ml of scintillation fluid (Hydromix, Yorktown Research) and counted in a Packard Tri-Carb 3375 liquid scintillation spectrometer with an efficiency of 33% for ^3H to calculate the total amount of specifically bound ^3H -dexamethasone, or analysed for total cellular protein by the method of Lowry¹⁵ using bovine serum albumin as a standard. Inset, Scatchard plots¹⁶ of the binding data for ^3H -dexamethasone by suspensions of A/J (○) or C57BL/6J (●) maxillary mesenchyme cells.

primary mesenchyme cells (maxillary) was measured as a function of the free steroid concentration. The curves indicate the presence of specific corticosteroid receptors, for dexamethasone in both cell types, which approach saturation between 5×10^{-9} M and 2.5×10^{-8} M dexamethasone. Scatchard plots of the data (inset, Fig. 1) are linear, indicating the presence of a single class of binding sites for dexamethasone in both A/J and C57 mesenchyme cells with an apparent K_D for dexamethasone of $\sim 8.5 \times 10^{-9}$ M and 1.3×10^{-8} M for A/J and C57 cells, respectively; and with 121 fmol of ^3H -dexamethasone bound per mg total cell protein and 54 fmol per mg cell protein for the two cell types. Table 1 represents cumulative data obtained from Scatchard plots¹⁶ from several experiments in

which the specific binding of ^3H -dexamethasone was measured in cell suspensions derived from primary cultures of either palatal or maxillary mesenchyme cells obtained from A/J or C57 embryos. Irrespective of the origin of the mesenchyme cells (that is palatal or maxillary), the amount of dexamethasone-receptor activity (cytoplasmic and nuclear) was consistently higher in the mesenchyme cells of A/J than in the corresponding C57 cells ($\sim$ twofold). A/J or C57 mesenchyme cells derived from either the secondary palate or maxillary processes do not differ significantly in total protein per cell, in the distribution of protein in cytosol or nuclear fractions (Table 2), or in the amount of ^3H -dexamethasone which is distributed *in vitro* at 37°C between the cytoplasmic and nuclear fractions (33.1 compared with 39% in the nuclear fraction for A/J and C57 cells, respectively) in cultured or freshly trypsinised cells. Therefore, the difference in total amount of glucocorticoid-receptor proteins in A/J and C57 mesenchyme cells as measured by whole cell binding seems to be partially accounted for by an apparent difference in the amount of dexamethasone-receptor protein(s) in the cytosols of A/J and C57 cells (see also Table 3).

Although C57 mesenchyme cells possess on average a lower concentration of dexamethasone-receptor proteins on a per cell basis (7,200 sites per cell compared with 16,300 sites per cell for A/J, calculated from the cell numbers), the apparent K_D for dexamethasone of the C57 receptors is higher, $\sim 7.9 \times 10^{-9}$ M, than the corresponding affinity of the receptors in the A/J cells for dexamethasone, $\sim 1.7 \times 10^{-8}$ M. The reason for these differences is not entirely apparent although a similar difference in the affinity constant for dexamethasone and quantitative differences in either the number or distribution of cytoplasmic and/or nuclear glucocorticoid receptors have been reported in cultured murine lymphosarcoma and L929 fibroblast cells between steroid-sensitive and resistant clones^{13,17–19}.

To determine whether these quantitative differences in dexamethasone-receptor activity between cultured A/J and C57 mesenchyme cells are due to alterations in the cells' ability to respond differentially in culture to certain growth factors which could alter the cell content of receptor proteins²⁰, dexamethasone-binding activity was measured in cytosol fractions prepared from the maxillary processes of 14-d-old embryos (Table 3). Although the absolute amount of ^3H -dexamethasone bound per mg cytosol protein in both A/J and C57 maxillary cytosols was six times less than the amount of ^3H -dexamethasone which was bound per mg total cell protein using whole cell suspensions (average 315 fmol of bound ^3H -dexamethasone per mg cell protein and 150 fmol per mg cell protein for cultured A/J and C57 cells, respectively), the dexamethasone-receptor activity was twice as high in the A/J maxillary cytosols (54.5 fmol bound ^3H -dexamethasone per mg cytosol protein) than in the C57 maxillary cytosols (23 fmol bound per mg cytosol protein). But the K_D for dexamethasone of the A/J cytosol receptor(s) ($\sim 2.9 \times 10^{-8}$ M) and for the C57 cytosol receptor(s) ($\sim 8.9 \times 10^{-9}$ M) were similar to those values estimated by using suspensions of cultured mesenchyme cells. Likewise, when freshly dissociated (trypsinised) A/J maxillary mesenchyme cells are labelled *in vitro*, they bind approximately three times as much dexamethasone (54 fmol bound ^3H -dexamethasone per mg cell protein) than the corresponding C57 mesenchyme cells (18 fmol bound ^3H -dexamethasone per mg cell protein). Therefore, the apparent difference in the amount of dexamethasone-receptor protein(s) between A/J and C57 mesenchyme cells is evident in cells which have either been freshly dissociated or grown in culture for several days. These results suggest that there is little or no loss of this phenotypic trait (receptor concentration) in primary cultures of mesenchyme cells.

These results demonstrate that mouse mesenchyme cells derived from either the secondary palate or maxillary pro-

Table 1 Characteristics of dexamethasone receptors in cultured mouse facial mesenchyme cells

Mesenchyme cells	A/J	Strain	C57BL/6J
Secondary palate	$N = 3$ $K_D = 1.3 \times 10^{-8} \text{ M} (\pm 0.28)$ $r = 420 \text{ fmol per mg cell protein} (\pm 150)$ $= 34 \text{ fmol per } 10^6 \text{ cells}$ $\sim 20,500 \text{ sites per cell}$ $P = 76.4 \text{ } \mu\text{g per } 10^6 \text{ cells} (\pm 27.2)$	$N = 4$ $K_D = 6.4 \times 10^{-9} \text{ M} (\pm 1.6)$ $r = 188 \text{ fmol per mg cell protein} (\pm 80)$ $= 14 \text{ fmol per } 10^6 \text{ cells}$ $\sim 8,400 \text{ sites per cell}$	
Maxillary processes	$N = 4$ $K_D = 2.0 \times 10^{-8} \text{ M} (\pm 1.3)$ $r = 210 \text{ fmol per mg cell protein} (\pm 75)$ $= 20 \text{ fmol per } 10^6 \text{ cells}$ $\sim 12,000 \text{ sites per cell}$ $P = 90 \text{ } \mu\text{g per } 10^6 \text{ cells} (\pm 10.6)$	$N = 4$ $K_D = 9.3 \times 10^{-9} \text{ M} (\pm 0.38)$ $r = 110 \text{ fmol per mg cell protein} (\pm 30)$ $= 10 \text{ fmol per } 10^6 \text{ cells}$ $\sim 6,000 \text{ sites per cell}$	
Average	$N = 7$ $K_D = 1.7 \times 10^{-8} \text{ M} (\pm 0.7)$ $r = 315 \text{ fmol per mg cell protein} (\pm 100)$ $= 27 \text{ fmol per } 10^6 \text{ cells}$ $\sim 16,300 \text{ sites per cell}$	$N = 8$ $K_D = 7.9 \times 10^{-9} \text{ M} (\pm 0.38)$ $r = 150 \text{ fmol per mg cell protein} (\pm 40)$ $= 12 \text{ fmol per } 10^6 \text{ cells}$ $\sim 7,200 \text{ sites per cell}$	

Cumulative data from several different experiments (N , number of experiments) $\pm$ s.e.m. showing the specific binding of ^3H -dexamethasone to receptors in whole cell suspensions. Data were analysed by the method of Scatchard¹⁶ to calculate the concentration of receptor-steroid complex (r) in equilibrium conditions and the affinity of the receptors for dexamethasone, dissociation constant (K_D). The protein content (P) was determined by the method of Lowry¹⁵. The number of sites per cell was calculated from the cell number, which was determined using a Coulter counter.

cesses possess cytoplasmic dexamethasone-binding proteins of high affinity and limited capacity. Furthermore, facial mesenchyme cells obtained from day 14 A/J embryos have on average approximately double the amount of receptor proteins than do mesenchyme cells obtained from C57 embryos at equivalent stages. This biochemical difference might partially account for the variations observed *in vivo* in the ability of these two strains of mice to respond differentially to the teratogenic effect of glucocorticoids.

The cytoplasmic dexamethasone receptors from A/J and C57 mesenchyme cells have biological activity in that cell suspensions labelled *in vitro* with ^3H -dexamethasone accumulate specifically bound radioactive steroid in their nuclei, presumably as a result of nuclear translocation¹¹. The number of dexamethasone-receptor sites per cell as well as the percentage of ^3H -dexamethasone which is trans-

Table 3 Characteristics of dexamethasone receptors in cytosols of maxillary processes

Strain	A/J	C57BL/6J
$N = 2$	$K_D = 2.9 \times 10^{-8} \text{ M}$ (± 2.2) $r = 54.5 \text{ fmol per mg}$ cytosol protein (± 1.5)	$N = 3$ $K_D = 8.9 \times 10^{-9} \text{ M}$ (± 2.6) $r = 23 \text{ fmol per mg}$ cytosol protein (± 6.7)

Pooled maxillary processes obtained from day 14 embryos were homogenised in 2.0 ml of buffer composed on 10 mM Tris-HCl, pH 7.4, 1.5 mM EDTA and 0.5 mM dithiothreitol in a Kontes homogeniser (20–25 strokes) at 4 °C. After homogenisation, sucrose was added to a final concentration of 0.5 M. Homogenates were centrifuged at 100,000g for 60 min at 4 °C. Aliquots of the supernatants (cytosol fraction) were incubated at 4 °C for 2 h with 1–100 nM ^3H -dexamethasone in the absence or presence of an excess of unlabelled dexamethasone (10^{-5} M) to correct for nonspecific binding²¹. A suspension of dextran (0.5%, dextran T-70, Sigma) coated charcoal (5.0%, activated charcoal, Sigma) was then added to each assay to separate free from protein-bound steroid. The tubes were centrifuged at 1,500g for 15 min at 4 °C, and aliquots of the supernatants were taken to assess the specific binding of ^3H -dexamethasone to cytosol proteins. The values in the table represent the average data obtained from Scatchard plots for each experiment (N , number of experiments) $\pm$ s.e.m.

ferred into the nucleus in A/J and C57 is in agreement with the values reported for other steroid-responsive cells in culture^{18,21,22}. The presence of functional glucocorticoid receptors in both the palatal and surrounding facial mesenchyme cells favours the likelihood that these cells have the capacity to respond biologically to physiological levels of glucocorticoids. In fact, unmetabolised cortisol has been localised in the cells of the foetal maxilla of day 12 A/J embryos after *in vivo* administration of ^3H -cortisol²³.

Glucocorticoids are known to inhibit the growth of primary cultures of rat, mouse and chick embryonic fibroblasts^{24–26} as well as the multiplication of established mouse fibroblast lines such as L929 (ref. 27). Cortisone can also inhibit *in vivo* the proliferation of mesenchyme cells in the developing mouse secondary palate in A/J and H-Velaz mice^{28,29}. Experiments are in progress to determine whether the growth of primary cultures of mouse facial mesenchyme cells is altered *in vitro* by glucocorticoids.

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Table 2 Distribution of ^3H -dexamethasone binding in cytosol and nuclear fractions of mouse maxillary mesenchyme cells

Strain	Receptor activity	% Nuclear transfer
	Cytosol fraction	Nuclear fraction
A/J	97.7* 16†	20.6 15.2
		Average: 33.1(36.7)
C57BL/6J	160* 33†	103 20.9
		Average: 39 (32.1)

Whole cell suspensions from either freshly dissociated maxillary cells ($\sim 20 \times 10^6$ cells)* or primary mesenchyme cultures ($\sim 4 \times 10^6$ cells)† were incubated with 10 nM ^3H -dexamethasone in the absence or presence of unlabelled dexamethasone (10^{-5} M) at 37 °C for 40 min. The cells were then centrifuged, washed in ice-cold PBS, recentrifuged and resuspended in 1.0 ml of hypotonic buffer, composed of 10 mM Tris-HCl, pH 8.0, 1 mM MgCl_2 and 2 mM CaCl_2 ; and lysed by freeze-thawing. The lysates were separated into crude nuclear pellets and crude cytosol supernatants after centrifugation (200g, for 5 min at 4 °C). Aliquots of the latter were taken to assess the specific binding of ^3H -dexamethasone to cytosol fractions and to determine the protein content. The nuclear pellets were resuspended in 4.0 ml of ice-cold hypotonic buffer and centrifuged. The pellets were dissolved in 0.5 ml of distilled water and aliquots were taken to assess the specific binding of ^3H -dexamethasone in the nuclear fractions and to determine the protein contents. Receptor activity is expressed as fmol of specifically bound ^3H -dexamethasone per mg of cytosol or nuclear proteins. The percentage of nuclear transfer of the dexamethasone-receptor complex was determined according to Croce *et al.*²² using the following formula:

$$\% \text{ nuclear transfer} = \frac{\text{fmol per mg nuclear proteins}}{\text{fmol per mg nuclear proteins} + \text{fmol per mg cytosol proteins}} \times 100$$

Values in parentheses represent the percentage of total cellular protein in the nuclear fractions.

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Reactivity of normal and brachypod mouse limb mesenchymal cells with con A

SINCE plant lectins, such as concanavalin A (con A) and wheat germ agglutinin (WGA), bind to specific carbohydrate residues¹, they have been used to investigate changes in plasma membranes during the course of normal development by monitoring lectin-induced agglutinability²⁻⁶, the binding affinities of carbohydrate moieties of cell-surface components for radioactively-labelled lectins^{2,5,6}, and the distribution of binding sites using fluorescently conjugated lectins⁷. It has been proposed that agglutinability with con A and WGA varies with the stage of differentiation of a

particular cell type^{1,2,5,6,8}. It seems reasonable to assume, therefore, that modifications of cell-surface structure, such as changes in carbohydrate components or in their distribution on or in the plasma membranes, could result in the manifestation of an aberrant phenotype. Support for this notion comes from studies in which a reduced affinity for insulin and various plant lectins by liver cell plasma membranes was observed in young, hyperglycaemic mice⁹. Several studies on hereditary skeletal disorders¹⁰⁻¹² including brachypodism (*bp^H*) in mice^{13,14} have suggested that these anomalies originate at a time when the surface-mediated events of cell recognition and sorting-out are resulting in the formation of prechondrogenic blastemata¹⁵. Since rudiments derived from the postaxial region of the hindlimbs are the most severely affected in brachypodism^{13,14} con A agglutination and binding experiments were carried out on mesenchyme cells from this region in both normal (+/+) and mutant (*bp^H/bp^H*) embryos during early limb development. The results presented here show that differences occur over the 3-d test period supporting the contention that plasma membrane surfaces change during developments^{1-6,8}. These developmental modifications are, however, temporally different between the two genotypically different cells which may be related to the ultimate manifestation of the abnormal phenotype.

Postaxial mesenchyme cells of 11-d stage embryos of both genotypes exhibited a similar degree of agglutination (Table 1). Cells from 12-d stage normal embryos, however, showed a significant reduction in agglutination, whereas agglutination of the brachypod cells remained essentially unchanged. It is during this period that myogenic and chondrogenic processes commence in normal mouse limb development²². The decreased agglutination exhibited by 12-d stage normal cells is in agreement with the contention that, as differentiation proceeds, con A and WGA agglutinability decreases^{1,8}.

A reduction similar to that observed in normal cells between the 11- and 12-d stages was not found until 24 h later

Table 1 Comparison of con A binding and agglutination of normal and brachypod limb mesoblast cells from 11-, 12-, and 13-d stage mouse embryos*

Stage and genotype	Percentage agglutination† (mean ± s.d.)	³ H-con A binding‡§ (c.p.m. / mg protein × 10 ⁻³)	Specific activity <i>bp^H/bp^H</i> / Specific activity +/+	Surface area (mm ²) (mean ± s.d.)
11-d				
+/+	45.4 ± 9.3	440		0.118 ± 0.030
<i>bp^H/bp^H</i>	40.9 ± 12.5	470	1.07	0.112 ± 0.029
12-d				
+/+	17.9 ± 10.2	210		0.123 ± 0.036
<i>bp^H/bp^H</i>	44.7 ± 13.7¶	540	2.57	0.118 ± 0.032
+/+ (trypsinised)	54.5 ± 4.7	400		
<i>bp^H/bp^H</i> (trypsinised)	56.2 ± 9.1	390	0.98	
13-d				
+/+	6.2 ± 2.7	390		0.108 ± 0.030
<i>bp^H/bp^H</i>	17.6 ± 10.9	280	0.72	0.110 ± 0.036

* Mutant and normal mesoblast cells from the postaxial region of the hindlimbs of 11-, 12-, and 13-d stage embryos were isolated as described previously¹⁶. After a 30-min treatment with Ca²⁺ and Mg²⁺-free (CMF) Tyrode's solution, dissociation was carried out in phosphate-buffered saline (PBS)¹⁷ containing 10 µg ml⁻¹ DNase³. In certain experiments, CMF treatment was replaced by a 20-min treatment with 1.5% trypsin at room temperature. Both types of dissociation treatments yielded cell suspensions of greater than 92% viability as determined by Trypan blue exclusion. Viability remained the same at the end of the experiments. Binding and agglutination experiments were carried out in the following conditions: a 0.4-ml reaction mixture containing 150 µg ml⁻¹ con A (Sigma) and 5.5 × 10⁵ cells in PBS-DNase solution was incubated in Falcon multiwell culture dishes for 20 min at 4 °C with occasional, gentle swirling. Controls for each genotype included 0.1 M α-D-methylmannose (Sigma) to inhibit lectin-induced agglutination. Aliquots of each reaction mixture were placed in depression slides and the percentage of total cells agglutinated (two or more cells in clumps) was determined by counting single cells against agglutinated cells in a minimum of six fields of view¹⁷. Statistical significance of the differences was determined using a two-group comparison *t* test¹⁸. Tritiated con A (New England Nuclear, 65.2 Ci mmol⁻¹) diluted with cold con A to a concentration of 150 µg ml⁻¹ was used for the binding experiments. Labelled cells were collected as described by Noonan and Burger¹⁹ and solubilised in Protosol (New England Nuclear) before counting in Omnifluor-toluene scintillation cocktail with a Beckman LS-100 scintillation counter. Protein determinations were carried out using Dulley and Grieve's modification of the Lowry procedure^{20,21}.

† Expressed as percentage agglutination over that found in controls.

‡ Expressed as c.p.m. × 10⁻³ above what was determined for the controls.

§ Average value for at least three determinations.

¶ CMF-treated, 12-d *bp^H/bp^H* significantly different from CMF-treated, 12-d +/+ but did not differ significantly from 12-d trypsinised +/+ or *bp^H/bp^H* (two-group comparison *t* test, *P* = 0.01).

in the mutant cells (Table 1). This delayed decrease in agglutination corresponds with the delay in the ability of mutant cells to produce matrix components and stain meta-chromatically in the intact limb^{13,14}, in monolayer cultures¹⁶, and in rotation-reaggregation cultures²³. It has been shown in other developing systems that the decrease in con A-induced agglutination is not accompanied by a concomitant drop in the binding of the lectin^{2,5}, suggesting that the decrease in agglutinability is due to the inability of the binding sites to rearrange into a display conducive to agglutination¹. This does not, however, seem to apply to this system (Table 1). There is a decrease in the amount of con A bound by the +/+ cells between days 11 and 12, whereas the mutants remain essentially the same. Comparing the specific activities of con A bound to bp^H/bp^H and that of +/+ at day 11, both genotypes bind approximately the same amount of lectin. The decline in the binding of con A by the +/+ cells at day 12 results in an increase in the ratio of specific activities, suggesting that twice as many binding sites are in bp^H/bp^H cells than in +/+ cells of the same gestational age. At day 13 the amount of lectin bound to bp^H/bp^H cells again approaches the normal condition. It is unlikely that the variations in binding can be explained solely by changes in surface area since this remained essentially unchanged in the two genotypes over the 3 d tested (Table 1).

The question remains as to whether the decrease in binding exhibited by the +/+ cells at day 12 is the result of loss of sites or masking of sites. Studies with trypsinised cells of both genotypes at day 12 (Table 1) supports the latter contention since the amount of tritiated lectin bound is the same with both trypsinised cell types. Enzyme-treated +/+ cells also exhibit the same degree of agglutination as trypsinised and non-trypsinised bp^H/bp^H cells. This supports the once held notion that trypsinisation unmasks "cryptic" sites²⁴ and suggests that 12-d stage +/+ and bp^H/bp^H cells possess the same number of con A-binding sites.

It is not understood at this time what the relationship is between the degree of agglutination, con A-binding sites, and the effect of the bp^H mutation. Work is presently in progress to determine agglutination with other plant lectins (WGA, fucose-binding protein) as well as the distribution of their respective binding sites using fluorescent conjugated lectins. Also, investigations using phenotypically normal bp^H /+ offspring are being carried out to determine the degree of functionality of the bp^H allele in the heterozygous condition.

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Genetic differences in susceptibility of pancreatic β cells to virus-induced diabetes mellitus

INFECTION of certain inbred strains of mice with the M variant of encephalomyocarditis (EMC) virus produces β -cell damage and a diabetes-like syndrome^{1–4}. The development of this syndrome is genetically determined and susceptibility is inherited as an autosomal recessive trait^{5,6}. The genetic factors involved seem to act at the level of the β cells, and *in vitro* studies have shown that β cells from susceptible strains of mice are more permissive to EMC infection than β cells from resistant strains of mice⁷. We have investigated first, the capacity of EMC virus to replicate in β cells from F₁ and F₂ offspring of susceptible and resistant strains of mice, and second, the effect of the viral infection on the level of insulin and glucose in the blood.

Four-to-five-week-old mice susceptible (SWR/J) and resistant (C57BL/6J) to the development of EMC-induced diabetes^{5,6}, as well as the F₁ and F₂ offspring, were infected intraperitoneally with 5.0×10^3 plaque-forming units (PFUs) of the M variant of EMC virus. At different times after infection, the pancreas was removed and β cells were isolated from individual mice as before⁷. In brief, the pancreas was minced, treated with collagenase (3 mg ml⁻¹ for 15 min at 37°C) and approximately 100 islets were collected from each pancreas with a stereo microscope. The islet-containing material was again treated with collagenase (7 mg ml⁻¹ for 6 min at 37°C), filtered through sterile gauze to remove large aggregates, and then layered on a 13–36% preformed Ficoll density gradient. The heavy visible band containing approximately 6.0×10^4 cells was collected⁷. Staining with aldehyde-thionin or fluorescein isothiocyanate-labelled anti-insulin antibody showed that almost 85% of the cells were β cells. These cells were then washed, frozen and thawed three times, homogenised and assayed for infectious virus on confluent monolayers of secondary mouse embryo cells. The concentration of insulin in the plasma was measured by the double antibody radioimmunoassay technique⁸ using mouse insulin (Novo A/S, Copenhagen) as the standard. Blood glucose levels were determined enzymatically by the glucose oxidase method with *o*-dianisidine dihydrochloride as the reactive dye⁹.

Figure 1 shows the relationship between insulin and glucose levels in the plasma at different times after infection. Before infection, the mean blood glucose level was 147 ± 19 mg per 100 ml with none of the glucose values above 182 mg per 100 ml or below 103 mg per 100 ml. The mean insulin level was 3.0 ± 0.95 ng ml⁻¹ with none of the insulin values below 1.9 ng ml⁻¹ or above 5.7 ng ml⁻¹. Within 48 h after infection, almost two-thirds of the SWR/J mice had insulin and glucose values outside these ranges. At 7 d, and even more clearly at 14 d, many of these mice became hypoinsulinaemic and hyperglycaemic. In contrast to the SWR/J mice, the glucose and insulin values of the C57BL/6J and F₁ hybrids generally fell within the normal range. Occasionally, 2 d after infection, a small percentage of the F₁ hybrids showed signs of hyperinsulinaemia, but this generally was not followed by hypoinsulinaemia. Several F₂ mice also showed signs of hyperinsulinaemia and hypoglycaemia 2 d after infection, but in contrast to the F₁ hybrids, at 7 and 14 d they became segregated into two groups—one with glucose and insulin levels within the normal range and the other showing severe hypoinsulinaemia and hyperglycaemia.

The viral titre in β cells isolated from mice at different times after infection is illustrated in Fig. 2. Data from 290 individual mice show that the greatest difference between SWR/J and C57BL/6J

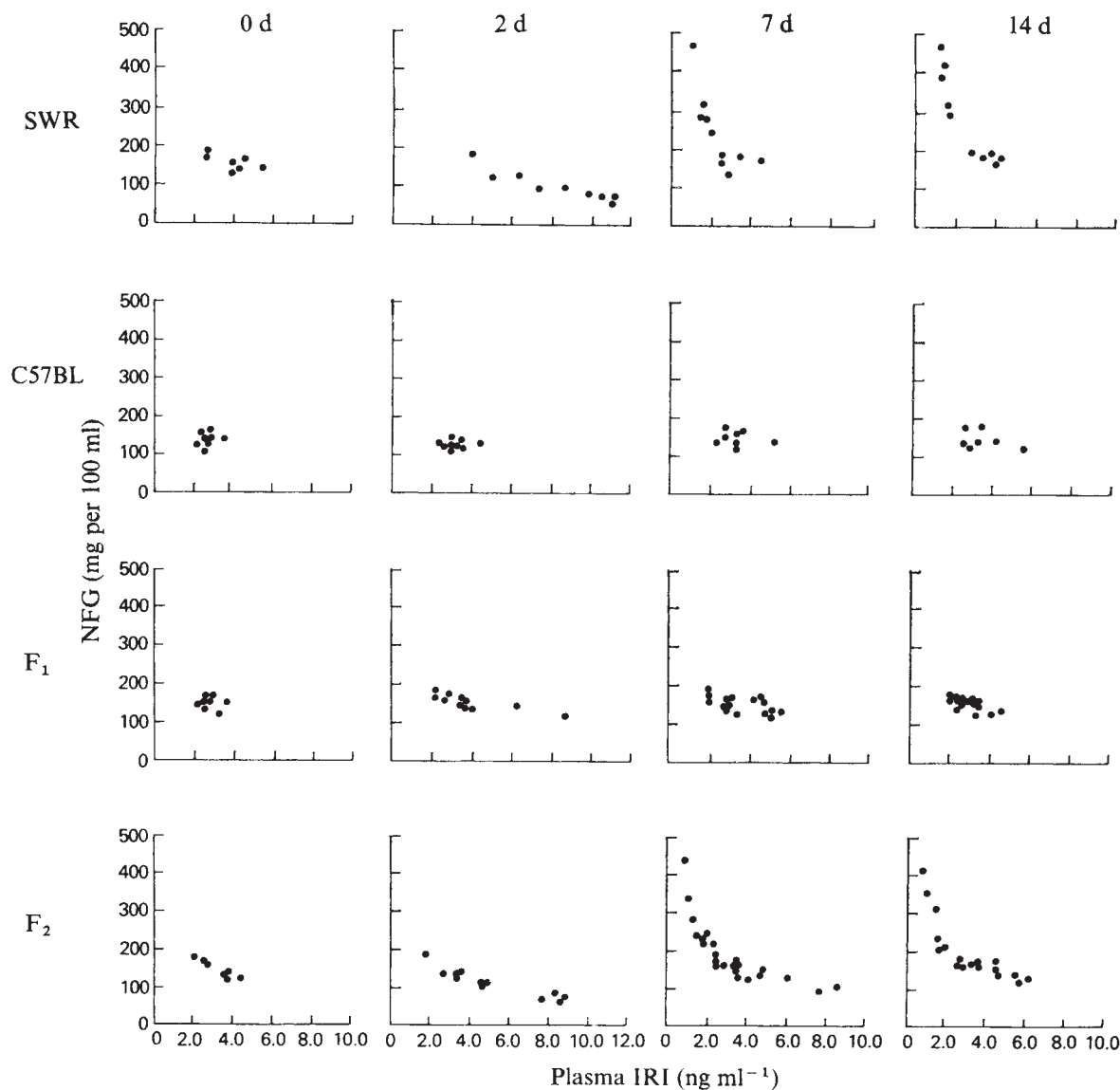


Fig. 1 Concentration of non-fasting glucose (NFG) and immunoreactive insulin (IRI) in the plasma of susceptible (SWR/J) and resistant (C57BL/6J) mice and their F_1 and F_2 offspring at different times after infection with EMC virus. Only male mice were tested and each point represents data from an individual animal.

occurred 48 h after infection and was more pronounced in males than females. At 48 h, up to 12 times more virus was recovered from β cells of SWR/J male mice ($2.6 \pm 2.8 \times 10^5$ PFUs) than C57BL/6J male mice ($2.1 \pm 0.7 \times 10^4$ PFUs), and analysis of variance revealed that the mean difference between the two groups was highly significant ($P \leq 0.005$). The mean virus titre in the β cells of the F_1 hybrids ($2.9 \pm 0.7 \times 10^4$ PFUs) was not significantly different from that of the resistant C57BL/6J mice ($P \geq 0.5$). The virus titre of individual F_2 animals, however, was spread out over a wider range and the standard deviation ($4.7 \pm 4.3 \times 10^4$ PFUs) was significantly different ($P < 0.005$) from that of the F_1 hybrids or the resistant C57BL/6J mice. The standard deviation of the F_2 females at 48 h also was significantly different ($P < 0.05$) from that of the female F_1 hybrids and the female C57BL/6J. Previous studies showed that male mice generally develop more severe hyperglycaemia than female mice after infection with EMC virus^{5,6}.

Figure 3 shows the relationship between viral titre and the levels of insulin and glucose in individual animals 48 h after infection. Although there were some exceptions, mice with the highest viral titre in the pancreas had the highest insulin and lowest glucose levels in the blood. Most of the SWR/J mice had viral titres

above 10^5 PFUs and their glucose and insulin values fell outside the normal range. In contrast, the resistant C57BL/6J and F_1 hybrid animals had viral titres below 10^5 PFUs and their glucose and insulin levels fell within the normal range. Animals in the F_2 generation segregated into two groups—the mice with viral titres above 10^5 PFUs had abnormal glucose and insulin values, while those with viral titres below 10^5 PFUs had glucose and insulin values within the normal range. Earlier studies had shown that the viral titres in the pancreas of susceptible mice was highest 2 d after infection and with the appearance of neutralising antibody at 3–4 d, the viral titre decreased^{5,7}.

We have shown that β cells from SWR/J mice are more susceptible to infection than β cells from C57BL/6J mice⁷. The experiments with individual mice reported here extend those observations, and show that the virus titre in β cells of F_1 hybrids was similar to that found in β cells of resistant C57BL/6J animals. Moreover, analysis of individual F_2 animals revealed two groups—one with viral titres approaching the susceptible SWR/J mice and the other with titres similar to that of the resistant C57BL/6J mice. Since the method of isolating β cells yielded preparations that were only about 85% pure⁷, the presence of virus

in the contaminating non- β -cell population might have obscured even greater differences in the actual viral titre in susceptible compared with resistant β cells. Nonetheless, our data show a correlation between viral replication and the subsequent development of clinical diabetes. Virus replicated best in β cells from SWR/J mice, and it was these animals that developed the most severe diabetes. Considerably less virus was found in β cells from C57BL/6J and F_1 hybrid mice, and these animals with high viral titres developed clinical diabetes. In contrast, the F_2 offspring segregated into groups with high and low viral titres; the animals with high viral titres were hyperinsulinaemic and hypoglycaemic at 2 d after infection due to acute β -cell damage and the rapid release of insulin. At about 7 d after infection these animals became hypoinsulinaemic and hyperglycaemic as a result of residual β -cell damage and the decrease in the insulin content of the pancreas^{1-4,7}. Because of the small size of the groups, the number of genes influencing susceptibility to virus-induced diabetes could not be analysed. Previous studies suggested that more than one gene was involved⁶.

Taken together, our studies suggest that EMC-induced hypoinsulinaemia and hyperglycaemia are secondary to genetically determined differences in the permissiveness of β cells to support viral replication. The demonstration of an association between particular HLA antigens (for example, B8 and BW15) and the frequency of juvenile diabetes supports the concept of a genetic predisposition to this disease¹⁰⁻¹⁴. The possibility that individuals

Fig. 2 Titre of virus in β cells from susceptible (SWR/J) and resistant (C57BL/6J) mice and their F_1 and F_2 offspring at different times after infection. Animals in the F_1 group represent both (SWR/J $\times$ C57BL/6J) F_1 and (C57BL/6J $\times$ SWR/J) F_1 . F_2 animals were bred from both (SWR/J $\times$ C57BL/6J) F_1 and (C57BL/6J $\times$ SWR/J) F_1 . Animals were infected intraperitoneally with 5.0×10^3 PFUs of EMC virus. The pancreas was removed, islets were collected and β cells were isolated by Ficoll density gradient centrifugation. The β cells were then homogenised and assayed for infectious virus on secondary mouse embryo cells. *a*, Male mice 24 h after infection; *b*, male mice 48 h after infection; *c*, male mice 72 h after infection; *d*, female mice 48 h after infection. Each point represents an individual animal and viral titre is expressed as PFUs per pancreas. Bars represent the geometric mean titre.

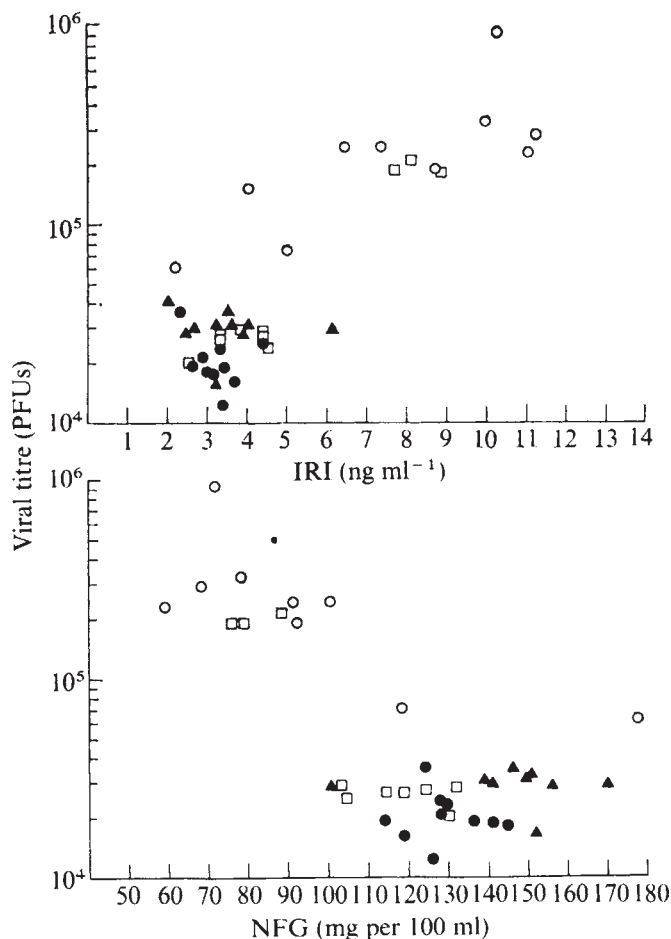
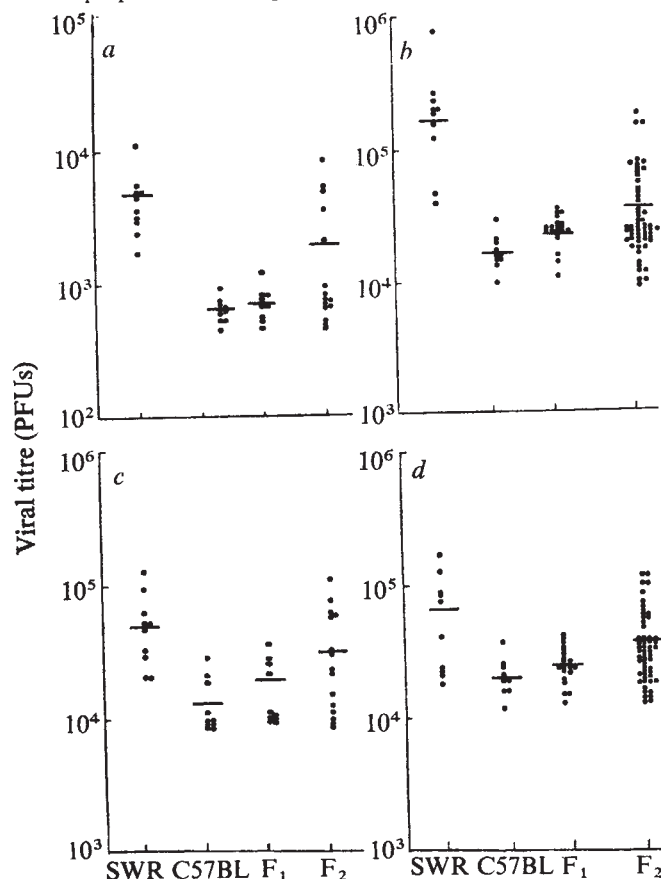


Fig. 3 Relationship between viral titre in β cells of the pancreas and level of insulin and glucose in the plasma of male mice 48 h after infection with EMC virus. See legend to Figs 1 and 2. $\circ$, SWR/J; $\bullet$, C57BL/6J; Δ , F_1 hybrids; $\square$, F_2 . Each point represents an individual animal.

of certain HLA types are more susceptible or respond differently to viral infections merits investigation.

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Activity of albumin conjugates of 5-fluorodeoxyuridine and cytosine arabinoside on poxviruses as a lysosomotropic approach to antiviral chemotherapy

THE therapeutic use of inhibitors of DNA synthesis as antiviral agents is hindered by their toxicity to dividing cells, which precludes the administration of effective doses. Consequently, although these drugs are very active in inhibiting virus replication *in vitro*, most of them are ineffective *in vivo*^{1,2}.

Albumin conjugates of two of these compounds, 5-fluorodeoxyuridine (FUdR)^{3,4} and cytosine arabinoside (ara-C) (G. B.-B. *et al.*, unpublished), release inhibiting substances in active form after penetration into the cells. This finding suggests a possible chemotherapeutic approach to attacking those DNA viruses which replicate in cells with a high protein uptake, that is, cells of the macrophage system⁵. After administration of such conjugates *in vivo* the inhibitors should be selectively concentrated in macrophages which are resistant to these drugs because they do not divide⁵. An attempt to concentrate in macrophages FUdR and ara-C by using their protein conjugates was suggested by the observation that the toxins amanitin and phalloidin after conjugation to albumin, change their original target selectively damage cells endowed with a high pinocytotic activity⁶⁻⁹.

Particularly suitable candidates to test this model of anti-

viral chemotherapy are poxviruses whose infectious cycle *in vivo* begins in macrophages¹⁰. We have studied the action of albumin conjugates of FUdR and ara-C: (1) on the replication *in vitro* of *Vaccinia* and *Ectromelia* viruses in cultures of L-929 cells and of mouse peritoneal macrophages; (2) on the growth of *Ectromelia* virus in mouse liver. *Ectromelia* virus, the agent of mousepox, is a poxvirus which, when injected intravenously into mice, is ingested mainly by the Kupffer cells in the liver where it starts replication. Then it infects neighbouring hepatic cells which in turn infect more hepatic cells after each cycle of growth. The resulting liver necrosis causes the death of mice^{11,12}.

Ectromelia virus (Hampstead mouse strain), grown in L-929 cells, was purified by zonal centrifugation in sucrose gradients according to Joklik¹³ and to Planterose *et al.*¹⁴ with slight modifications. The intravenous LD₅₀ of the purified virus for Swiss male mice of 28–30 g was 6.7×10^2 plaque forming units per g body weight. The *Vaccinia* virus used was from crude stocks produced in L-929 cells. The preparation of the FUdR-albumin conjugate (FUdR-RSA I) was carried out as described previously⁴ by coupling FUdR to crystalline rabbit serum albumin (RSA) by means of 1-ethyl-3-(dimethyl-aminopropyl)carbodiimide (EDCI) (Fluka). Ara-C-albumin conjugate (ara-C-RSA) was prepared by using the same concentration of reactants and by following the same procedure used for FUdR-RSA I. The molar ratios of FUdR and ara-C to RSA in the conjugates, determined in separate preparations by using ³H-FUdR and ³H-ara-C (Radiochemical Centre), were found to be 30 and 18 respectively. Sodium dodecyl sulphate (SDS)-polyacryla-

Table 1 Effect *in vitro* of free and conjugated FUdR and ara-C on *Ectromelia* and *Vaccinia* virus replication

Experiment	Compounds	$\mu\text{g ml}^{-1}$	Virus yield (log ₁₀ PFU ml ⁻¹)	
			<i>Ectromelia</i>	<i>Vaccinia</i>
1	Control		2.49	4.26
	FUdR	2.5	1.38 (45)†	1.72 (60)†
	FUdR-RSA I	25 (2.6)*	1.06 (57)	3.48 (19)
	FUdR-RSA I	50 (5.3)	0.80 (68)	1.36 (68)
2	Control		2.45	4.31
	FUdR	2.5	0.56 (77)	1.96 (55)
	FUdR-RSA II	25	1.23 (50)	2.74 (37)
	FUdR-RSA II	50	0.38 (84)	1.60 (63)
	EDCI-RSA	50	2.65 (0)	4.38 (0)
3	Control		2.29	4.22
	ara-C	2.5	0 (100)	0 (100)
	ara-C-RSA	25 (1.5)	1.58 (31)	2.13 (50)
	ara-C-RSA	50 (3.1)	0.38 (83)	1.42 (66)
4	Control		—	3.81
	FUdR-RSA I	50 (5.3)	—	1.39 (64)
	FUdR-RSA I	50 (5.3)	—	—
	+ thymidine	100	—	3.91 (0)
	FUdR-RSA II	50	—	1.51 (60)
	FUdR-RSA II	50	—	—
	+ thymidine	100	—	3.59 (6)
	ara-C-RSA	50 (3.1)	—	1.36 (64)
	ara-C-RSA	50 (3.1)	—	—
	+ deoxycytidine	50	—	2.69 (29)
	ara-C-RSA	50 (3.1)	—	—
	+ deoxycytidine	500	—	2.99 (22)
5	Control		1.25	
	FUdR	7	0.25 (80)	
	FUdR-RSA I	50 (5.3)	0.38 (70)	
	FUdR-RSA II	100	0.87 (30)	

Cells were infected with *Ectromelia* or *Vaccinia* virus at the input multiplicity of 1 PFU per cell. Virus titres were determined by plaque counting in L-929 cells according to Blanden³¹ with an overlay medium containing 0.5% methylcellulose. Virus yields were obtained by subtracting the amount of virus adsorbed to cells from the virus titre at the end of the experiment. Mouse peritoneal macrophages were collected and cultured as described previously⁷. Experiments 1–4 were performed in L-929 cells, experiment 5 in mouse peritoneal macrophages.

*The amounts (μg) of inhibitors contained in the conjugates are shown in parentheses.

†Number in parentheses is the percentage of inhibition.

mid gel electrophoresis showed that only a small portion of the conjugates had a molecular weight corresponding to the monomeric form of albumin. The bulk of the conjugates was made up by molecular species corresponding to high molecular weight polymers of albumin which were formed in the presence of ECDI¹⁵. Since in ECDI conjugation stable covalent bonds are also formed between albumin and the urea derivative of carbodiimide¹⁵, RSA was allowed to interact with ECDI in the absence of FUDR or ara-C so that we could evaluate the possible antiviral activity of this complex (ECDI-RSA). We also studied the antiviral activity of another FUDR-RSA conjugate prepared by Dr de Vries in the laboratory of Professor Th. Wieland (Heidelberg). This conjugate (FUDR-RSA II) was obtained by coupling the hydroxysuccinimide ester of succinylated FUDR to RSA. This conjugation produces neither side reactions nor polymerisation of albumin¹⁶ and consequently avoids the heavy changes of protein molecules which follow carbodiimide coupling reactions. The molar ratio of FUDR to RSA in this conjugate was not known.

Table 1 shows that free and conjugated FUDR and ara-C inhibit the replication of *Ectromelia* and *Vaccinia* viruses *in vitro* after infection of L-929 cells or mouse peritoneal macrophages. *Ectromelia* virus yields in mouse peritoneal macrophages are lower than those in L-929 cells since virus replication is restricted in the former cells¹⁷. FUDR-RSA I and II are equally active *in vitro* on both viruses; ECDI-RSA has no effect. Thymidine and deoxycytidine, which are known respectively to remove totally or partially the effects of FUDR and ara-C¹⁸, counteract the antiviral activity of FUDR-RSA and of ara-C-RSA, indicating that the activity of the conjugates is due to the FUDR and ara-C moieties. Since albumin after penetration into the cells by endocytosis is broken down by lysosomal enzymes¹⁹, it is likely that the conjugates exert their antiviral activity after digestion of the protein moiety and release of the drugs, thus behaving as lysosomotropic agents^{20,21}. The assumption that the bonds linking FUDR and ara-C to RSA are broken after penetration of the conjugates into the cells is supported by the evidence that these bonds involve the primary hydroxyl

groups of FUDR and ara-C²² which must be phosphorylated so that the drugs can block DNA synthesis¹⁸.

The effects of FUDR-RSA and ara-C-RSA *in vivo* have been evaluated according to two criteria: (1) inhibition of virus production in liver; (2) survival time and number of survivors. The results of the experiments on virus production in mouse liver are given in Table 2. The compounds were injected intravenously. FUDR-RSA I, FUDR and ara-C were given at doses which, with the time schedule followed, corresponded to 1/3, 1/2 and 1/3 of the respective LD₅₀. Ara-C-RSA administered to normal mice with the schedule of experiment 3 but in doses three times higher did not kill any of the injected animals. Experiments 1-5 show that FUDR-RSA I and ara-C-RSA reduced significantly the virus yield in mouse liver whereas free FUDR and ara-C as well as ECDI-RSA were ineffective. Note that the active doses of the conjugates contained 19 and 15 times less FUDR and ara-C respectively compared with the amounts of free compounds used. FUDR-RSA II, which *in vitro* was as active as FUDR-RSA I was ineffective *in vitro* (experiment 4). The inactivity *in vivo* of FUDR-RSA II may depend on lack of aggregation and of profound protein changes, allowing the conjugate to escape a rapid uptake by macrophages²³⁻²⁵. In contrast, an aggregated and heavily modified albumin such as FUDR-RSA I is very quickly removed from the blood and taken up mainly by the Kupffer cells in the liver²³⁻²⁶. No reduction of virus titre by ara-C-RSA, determined in experiment 4, was found in the spleens. This result agrees with the observation that in mice the uptake of aggregated albumin is several times lower in spleen than in liver²³. Experiment 6 shows that ara-C-RSA is slightly active in reducing virus titre in the liver if its administration starts 12 h after infection, when the cycle of virus replication in Kupffer cells is completed^{11,12}. This finding demonstrates that the conjugates interfere mainly with the phase of virus infection which takes place in liver macrophages. This is supported by the results of experiment 7 showing that ara-C-RSA reduces the virus titre also if administered only within the first 12 h of infection. Table 3 shows the results of the survival experi-

Table 2 Effect *in vivo* of free and conjugated FUDR and ara-C on *Ectromelia* virus replication in mouse liver

Experiment	Compounds	Doses*	Virus yield (log ₁₀ PFU ml ⁻¹)
1	Controls		7.7
	FUDR-RSA I	100(0)	6.0
	ECDI-RSA	100(0)	7.8
2	Controls		7.6 ± 0.1
	FUDR	200(0)	7.3 ± 0.2 (NS)
	FUDR-RSA I	150(0)	6.3 ± 0.2 P < 0.001
3	Controls		7.4 ± 0.1
	ara-C	70(0)	7.3 ± 0.4 (NS)
	ara-C-RSA	150(0)	5.6 ± 0.8 P < 0.005
4	Controls		7.5 ± 0.3
	ara-C-RSA	75(0)	6.2 ± 0.6 P < 0.01
	ara-C-RSA	37(0)	7.2 ± 0.2 (NS)
	FUDR-RSA II	150(0)	7.4 ± 0.4 (NS)
5	Controls		7.5 ± 0.6
	ECDI-RSA	150(0)	7.8 ± 0.8 (NS)
6	Controls		7.8 ± 0.2
	ara-C-RSA	150(0)	6.1 ± 0.5 P < 0.001
	ara-C-RSA	150(12)	7.3 ± 0.1 P < 0.005
7	Controls		7.8 ± 0.5
	ara-C-RSA	150(0)	6.4 ± 0.8 P < 0.01
	ara-C-RSA	150(0)	6.6 ± 0.4 P < 0.005

Purified *Ectromelia* virus was injected intravenously into mice at the multiplicity of 2.4×10^5 PFU per g body weight corresponding to 360 times the LD₅₀. 48 h after infection livers were collected and homogenised in 5 ml of Hanks' balanced salt solution. Virus titres of clarified supernatants were determined in L-929 cells³¹. Five animals were used in each group. The results are given as the mean ($\pm$ s.d.) of values obtained from livers of single animals, except in experiment 1 where livers from mice of each group were pooled before titration. The results were statistically evaluated by means of Student's *t* test and considered non significant (NS) when *P* was > 0.05.

*Doses are $\mu\text{g g}^{-1}$ body weight; in parentheses, h after infection at which compounds were injected intravenously.

Table 3 Effect of conjugates on survival of *Ectromelia* virus-infected mice

Experiment	Compounds	Doses*				Mean survival time (h)
1	Controls					72 ± 9
	FUdR-RSA I	150(0)	150(12)	50(24)	50(48)	65 ± 11 (NS)
	ara-C-RSA	150(0)	150(12)	50(24)	50(48)	94 ± 12 $P < 0.005$
No. survived/No. injected						
2	Controls					0/52
	ara-C	70(0)	70(12)	35(24)		0/7
	ara-C-RSA	150(0)	150(12)	50(24)		6/16 $P < 0.0005$
	ara-C-RSA	150(0)				5/24 $P < 0.01$
	ara-C-RSA	150(24)	150(36)	50(48)		0/12
	ECDI-RSA	150(0)	150(12)	50(24)		0/12

Experiment 1: mice were injected intravenously with the same amount of virus used in the experiments of Table 2. Twelve animals were used in each group. The results were evaluated statistically by means of Student's *t* test.

Experiment 2: mice were injected intravenously with 5.4×10^3 PFUs per g body weight, equivalent to 8 times the LD₅₀. Number of survivors was determined 21 d after infection. The results were evaluated statistically by means of the χ^2 analysis.

*For explanation of doses see Table 2.

ments. When mice were challenged with an amount of virus equivalent to 360 times the LD₅₀, ara-C-RSA increased significantly the mean survival time, whereas FUdR-RSA I was inactive. Since FUdR-RSA I reduces the virus titre *in vivo* to the same extent as ara-C-RSA, the different effect of the two conjugates on survival time is probably due to higher toxicity of the former. When mice were infected with eight times the LD₅₀ a significantly higher number of survivors was observed in the groups treated with ara-C-RSA than in the control group. Ara-C-RSA was active only if administered during the period of virus replication in Kupffer cells. Free ara-C and ECDI-RSA were ineffective.

In conclusion, albumin conjugates of FUdR and ara-C inhibit the growth of *Ectromelia* virus in mouse liver whereas the free inhibitors are ineffective. The finding that ara-C-RSA exerts its antiviral activity in liver macrophages indicates that ara-C is concentrated in these cells following the injection of the conjugate, providing experimental support to the lysosomotropic approach of virus chemotherapy. This observation also suggests that these or similar conjugates could be effective not only on viruses but also on other intracellular microorganisms growing in liver macrophages. In future experiments we will try to concentrate FUdR and ara-C in hepatocytes by use of a conjugate obtained by coupling these drugs to desialylated fetuin which is taken up specifically by parenchymal liver cells^{27,28} where it is digested by lysosomal enzymes²⁷. Such a conjugate should inhibit the replication of *Ectromelia* virus in hepatocytes. This line of research could have implications in the treatment of other hepatic viral infections such as human hepatitis B, whose agent has been localised in hepatocytes²⁹ and has been found to be a DNA virus³⁰.

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Experimental production of sheep pulmonary adenomatosis (Jaagsiekte)

SHEEP pulmonary adenomatosis (SPA) or Jaagsiekte is a contagious disease of unknown aetiology¹. A herpesvirus has been isolated from such tumours in Britain² and in Africa^{3,4} but attempts to produce adenomatosis with this virus have been unsuccessful (ref. 5 and W.B.M., G. Robinson, and K.W.A., in preparation). More recently, evidence has been presented that a type C virus with a density on a sucrose gradient of 1.5–1.20 g ml⁻¹, containing 60–70S RNA and RNA-directed DNA polymerase (reverse transcriptase) is present in the adenomatous tissue⁶. We wish to describe the production of SPA tumours in sheep experimentally infected with both agents.

Techniques⁷ similar to those of Perk *et al.*⁸, were used to confirm the presence of a reverse transcriptase-producing (RTP) agent in SPA tumour tissue from Scottish sheep. This enzyme was not detected in similar fractions from the lungs of unaffected sheep (Table 1).

A suspension of tumour from a natural case of SPA was subjected to equilibrium density centrifugation on a sucrose gradient. Fractions with densities of 1.15–1.17 g ml⁻¹ were centrifuged at 150,000g for 60 min at 4 °C and the pellet,

Table 1 DNA polymerase activity of fractions of tumour or unaffected lung

Sheep no.	Clinical state	Reagent added to standard reaction mixture		
		No addition	+ Poly(A)-oligo(dT) ₁₀	+ Actinomycin D
A525	Adenomatosis	60* (1)‡	930 (15)	70 (1.2)
J2	Adenomatosis	150 (1)	370 (2.5)	33 (0.2)
J4	Adenomatosis	40 (1)	330 (8.2)	26 (0.7)
J13	Adenomatosis	110 (1)	800 (7.3)	115 (1.1)
J14	Adenomatosis	153 (1)	750 (4.9)	133 (0.9)
N1	Unaffected	120 (1)	155 (1.3)	10 (0.09)
N15	Unaffected	117 (1)	173 (1.5)	47 (0.4)
N16	Unaffected	170 (1)	313 (1.8)	93 (0.6)

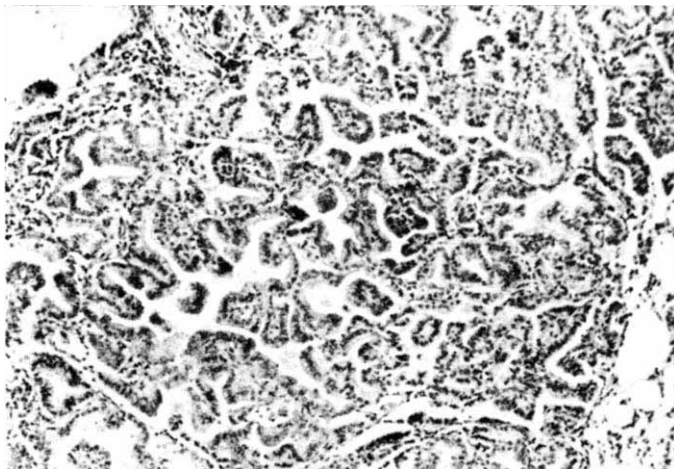
*Polymerase activity of 1.15–1.17 g ml⁻¹ fraction from sucrose density gradients of tumour or lung tissue suspensions, measured as increase in c.p.m. ³H-TTP incorporated into acid-insoluble material in 20 min using the methods outlined by Norval *et al.*

‡Ratio of comparative activities are given in parentheses and the discriminatory effect of poly(A)-oligo(dT)₁₀ and actinomycin D on polymerase activity in the adenomata and healthy tissues are shown.

resuspended in 199 medium, was injected endobronchially into six Cheviot sheep. Herpesvirus isolated from an SPA tumour and passaged four times in embryo sheep kidney cells and once in sheep pulmonary macrophages (titre $\geq 10^{6.7}$ TCD₅₀ ml⁻¹) was similarly injected into four sheep. Both the herpesvirus and the density gradient fraction, containing the RTP agent, were injected into six more sheep. As controls, four sheep were given a supernate of tumour suspension and four, from the same flock, a supernatant prepared in an identical manner from apparently normal lung. To avoid cross infection, the groups of sheep were housed, as in previous transmission experiments, in separate loose boxes.

Results are shown in Table 2. One sheep, given the tumour supernatant, was clinically affected with SPA 6 months after inoculation. This diagnosis was confirmed histologically and all sheep were killed therefore 7–8 months after inoculation. A minimum of ten standard sites from the lungs of each sheep were examined histologically. In the group given the RTP agent alone, one sheep had two small (3–5-mm) adenomata and one other, histological evidence of adenomatous changes. Lesions in both sheep

were minimal and present in four (13%) of a total of thirty-one sites examined. Of the sheep given both the RTP agent and the herpesvirus, three had definite macroscopic tumours and one was histologically positive (Fig. 1). No adenomatous lesions were found in any other sheep. All lungs were assayed for particles with reverse transcriptase activity and these were detected in lung tissue from two sheep (Table 2). Herpesvirus, which we have isolated from 24% of SPA tumours over the past 5 yr, was not recovered from any lung.

**Fig. 1** Focus of adenomatous change.

We can find no evidence to support the view that these tumours arose adventitiously or were caused by myoplasma or other cultivable microorganisms. Since some sheep in both of the groups given the density fraction containing the RTP agent developed adenomatous lung lesions within 8 months of inoculation, it seems certain that this agent was involved in the production of these changes. Whether it alone is capable of initiating adenomatous changes or requires the assistance of the herpesvirus is conjectural and remains to be established. In the four affected sheep given both herpesvirus and RTP agent, lesions were extensive, and present in a mean of 63% of the areas of lung examined. This may indicate that the herpesvirus was involved in the production of these changes. The possibility that a small number of herpesvirus particles was present in the fraction containing the RTP agent cannot be dismissed completely and such contamination would simulate, to a lesser extent, the combined inoculum of herpesvirus and RTP agent. It may be that both viruses act synergistically to cause oncogenic transformation of the target cells, the type II alveolar cells or the cells of Clara in the bronchioles*, but the precise role of these viruses can only be elucidated by further experiments with purified virus.

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Table 2 Evidence of pulmonary adenomata in the inoculated sheep

Material inoculated	No. in group	Macroscopic tumours	Histological adenomatous changes	Number with reverse transcriptase activity
Tumour supernatant	4	1	2	1
Herpesvirus and reverse transcriptase-producing agent	6	3	4	1
Reverse transcriptase-producing agent	6	1	2	0
Herpesvirus	6	0	0	0
Normal lung supernatant	4	0	0	0

Suspensions of finely minced tumour or lung tissue were prepared in Eagle's medium containing antibiotics and centrifuged at 12,000g for 30 min at 4 °C (inocula of "tumour or normal lung supernatant"). The supernate was recentrifuged at 120,000g for 90 min and the resulting pellet gently resuspended in 1 ml TNE buffer, layered on to a preformed gradient of 20–70% sucrose prepared in TNE buffer and centrifuged in a Beckman SW 40 rotor at 85,000g for 16 h at 4 °C. Fractions between densities of 1.15–1.17 g ml⁻¹ (the density of RNA tumour viruses) were pooled, diluted in TNE buffer and centrifuged at 150,000g for 60 min at 4 °C. The pellet obtained was suspended in 0.15 ml of TNE buffer and assayed for reverse transcriptase or further diluted for inoculation of sheep. Each inoculum was injected endobronchially in 8-ml volumes.

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Specific inhibition of mitochondrial Ca^{2+} transport by antibodies directed to the Ca^{2+} -binding glycoprotein

WE have described a glycoprotein isolated from various mitochondria¹, which binds Ca^{2+} with a dissociation constant of the same order of magnitude as the K_m for Ca^{2+} transport^{2,3}. This and other observations, including the effects of known Ca^{2+} transport inhibitors^{4,5} suggest that the protein is involved in the carrier mechanism for the transport of calcium in mitochondria. We have concluded that even passive efflux of Ca^{2+} from mitochondria requires the glycoprotein to be associated with the membrane⁶, and we consider this to be strong evidence that the passage of Ca^{2+} across the mitochondrial membrane requires the mediation of a transport system including the glycoprotein. We have sought to confirm these conclusions by immunological methods.

Rabbits were immunised using 2 mg of purified ox liver glycoprotein isolated in our laboratory as before⁷. Antigen plus Freund's complete adjuvant was given in five injections into all four legs and the neck. The operation was repeated with Freund's incomplete adjuvant a month later. A secondary immune response was evoked by intravenous injection of 0.2-0.5 mg of glycoprotein, and at weekly intervals 40 ml blood samples were taken from the marginal vein of the ear. The serum γ -globulin fraction was obtained by Keckwick's method⁸. Lipoproteins were removed by acidification with 0.05 M acetate buffer, pH 5.6.

Fig. 1 Elution pattern from Affi-Gel 10-glycoprotein column of γ -globulin fraction from rabbit serum. A sample of 4 mg of pure ox liver Ca^{2+} -binding glycoprotein taken up in 25 ml of 0.1 M bicarbonate-HCl buffer, pH 7, was added in 1 g of Affi-Gel 10 (BioRad). The mixture was stirred at 0 °C for 24 h. The resin was packed in a short column and washed free of 260 nm-absorbing material. 10 ml of 1 M ethanolamine was passed through the column and equilibration was carried out using 50 mM triethanolamine-HCl buffer, pH 7.8 containing 100 mM KCl. The sample was added in the same buffer.

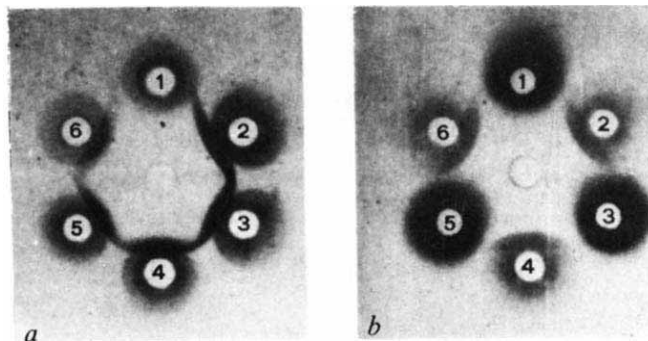
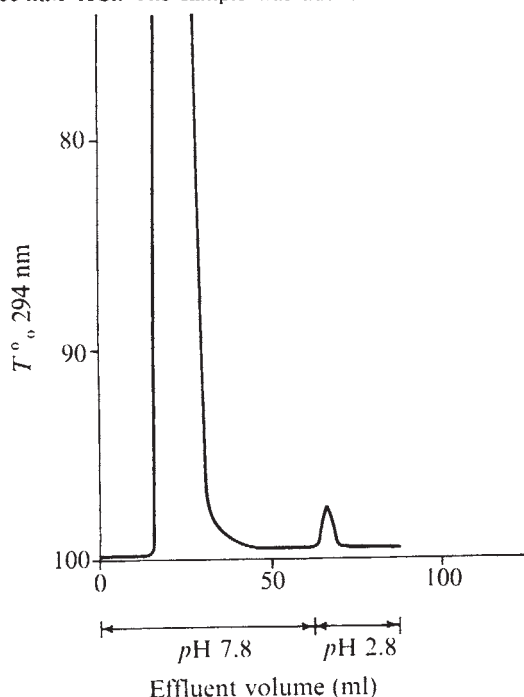


Fig. 2 Immunodiffusion patterns of γ -globulin fractions before (a) and after (b) affinity chromatography on Affi-Gel 10. See text for explanation.

Preliminary experiments with γ -globulin fractions, derived from the same animal before and after immunisation with the glycoprotein, had shown that γ globulins could to some extent inhibit Ca^{2+} transport in rat liver mitoplasts (mitochondria devoid of the outer membrane). To avoid non-specific effects, we purified the specific antibodies by affinity chromatography (Fig. 1). Only a small portion of the total protein was retained on the column and was eluted only when the pH was shifted from 7.8 to 2.8 (500 mM formic acid-ammonium formate). Since the limiting factor in this experiment is the amount of antigen coupled to the resin, the whole procedure was repeated four times. The last passage on the column yielded only negligible amounts of specific antibodies. We calculated that the overall purification was of the order of 150 times. Starting from about 1.5 g of serum protein, 10 mg of purified antibodies was obtained. Figure 2 shows the immunodiffusion patterns obtained by Ouchterlony's method⁹ by testing the γ -globulin fraction before and after immunisation with the glycoprotein. The central well (Fig. 2a) contained the isolated glycoprotein, well (1) contained the γ -globulin fraction before immunisation and all the other wells (2-6) contained γ -globulins obtained from the animal after secondary immunoresponses had been evoked at approximately 20-d intervals. A single antibody class was present in all samples. As a control (Fig. 2b) proteins after complete absorption on a column of Affi-Gel were tested against the glycoprotein. They did not contain appreciable amounts of antibodies (wells (1), (3) and (5)). Immunoprecipitate was present in wells (2), (4) and (6), where the γ -globulin fraction had been placed before affinity chromatography.

Figure 3 shows the inhibitory activity of the purified antibodies on Ca^{2+} transport by mitoplasts and intact mitochondria. Ca^{2+} transport by mitoplasts showed a hyperbolic dependence on antibody concentration, as if the antibodies were titrating specific sites freely accessible to them. As expected, on the other hand, mitochondria were much less sensitive to added antibodies, and decayed linearly as antibody concentration increased. Half inhibition was attained at approximately 1.5 μg antibody protein per mg protein with mitoplasts. The same effect was obtained in mitochondria with an antibody concentration about five times greater. It could be argued that antibodies against a mito-

Table 1 Effect of specific antibodies to the Ca^{2+} -binding glycoprotein on Ca^{2+} transport, electron transport and respiratory control ratios in isolated rat liver mitoplasts

Antibody added (μg)	% Inhibition		
	Ca^{2+} transport	Electron transport	RC
5	45	0	0
20	70	17	10
40	84	37	19

A Clark oxygen electrode was used at 30 °C (ref. 13). The substrate succinate and reaction mixture were as described before¹⁰. RC, Respiratory control.

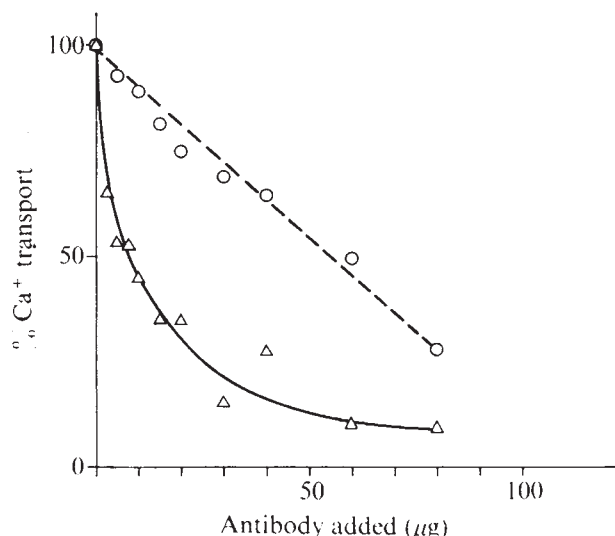


Fig. 3 Effect of antibodies against the glycoprotein on Ca^{2+} transport in rat liver mitochondria and mitoplasts. Mitochondria and mitoplasts were prepared as before¹⁰; Ca^{2+} transport was followed by the murexide method¹¹ in a mixture containing in 3 ml: 5 mg mitochondria or mitoplast protein (Biuret reaction of Gornall *et al.*¹²), 100 μM murexide, 70 mM sucrose, 220 mM mannitol, 2 mM HEPES, pH 7.4, 5 mM acetate, 1 mM phosphate buffer, pH 7.4, 15 μM rotenone, 165 μM Ca^{2+} , 5 mM succinate. A 540–507 was recorded at room temperature with a Phoenix dual wavelength recording spectrophotometer. ○, Intact mitochondria; Δ, mitoplasts.

chondrial membrane component exert an inhibitory effect by inhibiting either electron transport or the energy conservation mechanism or both. Table 1 compares the effect of antibodies on Ca^{2+} transport, electron transport and respiratory control ratio in mitoplasts. Clearly low concentrations of antibody can block 45% of Ca^{2+} transport without effect on other mitochondrial activities. Even at higher concentrations the effect on electron transport and on respiratory control ratio was definitely lower than that on the movement of the cation.

These data indicate that specific blocking of the calcium-binding glycoprotein inhibits active Ca^{2+} transport. It is not surprising that the effect is much more evident in mitoplasts in view of the barrier present in intact mitochondria to the interaction between glycoprotein and antibody. On the other hand, intact mitochondria would be expected to be sensitive to the antibodies, in spite of the impermeability of the outer membrane to proteins. Preincubation for 1 h is necessary to obtain an almost complete combination between antigen and antibodies. During this time it is reasonable to assume that proteins could diffuse slowly across the outer mitochondrial membrane. This process has to be linearly correlated with the antibody concentration outside the mitochondria, which would explain the linear inhibition of Ca^{2+} transport in mitochondria. These data indicate that the mitochondrial calcium-binding glycoprotein is a necessary ingredient in mitochondrial Ca^{2+} transport.

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Possible involvement of thiamine in acetylcholine release

ALTHOUGH it has long been thought that thiamine (vitamin B₁) and its phosphoric acid esters might be linked directly to excitation and propagation of nerve action potentials^{1,2}, the relationship has not been established firmly^{3,4}. We wish to report results which suggest that involvement with synaptic transmission rather than axonal conduction might explain why vitamin B₁ is required for nervous system activity. Experiments were performed on the electric organ of *Torpedo marmorata*, a richly innervated cholinergic tissue⁵. The results indicated that thiamine and its phosphoric esters are involved, either directly or indirectly, in the release of the neurotransmitter acetylcholine (ACh).

A first approach to the problem was by comparing, in different conditions, ACh content with that of thiamine and thiamine phosphate esters of the electric organ (Table 1). Total thiamine content (thiamine plus esters) was found to range between 350 and 500 nmol g⁻¹ of wet weight, with a rather large variation from one *Torpedo* to another. The relative distribution of thiamine, thiamine monophosphate (TMP) and thiamine diphosphate (TPP) was different in the following conditions. (1) When the tissue was incubated in physiological medium, most of the compound was found in the form of phosphate esters and the total content of ACh could be extracted without loss. (2) Homogenisation in the same medium before extraction resulted in significant hydrolysis of TPP into thiamine and a 45% loss of ACh; this part of the total transmitter destroyed by homogenisation is the fraction called "free ACh"⁶. (3) After incubation in a medium with high Mg²⁺ and low Ca²⁺, which is known to block synaptic transmission, 33% of the esters were hydrolysed into thiamine and the amount of "free" ACh was decreased. (4) Continual stimulation of the electric organ at 5 per s, reported to induce sustained oscillations of the "free" ACh level^{7,8}, had the same effect on the phosphorylation of thiamine. In these preliminary experiments, however, the precise parameters of the thiamine oscillation were not established. We concluded that the level of "free" ACh in the electric organ was in some way related to the phosphorylation of thiamine.

In a second approach to the problem, electrophysiological and radiochemical methods were used to measure the effects of external thiamine and related compounds on synaptic transmissions. The first compound studied was oxythiamine, an analogue of thiamine known to inhibit the enzymes requiring TPP as cofactor but to have little or no effect on nerve excitation⁹. When added to the saline medium, oxythiamine caused considerable changes in the shape of single electrical discharges; both the amplitude and duration increased (Fig. 1). This was undoubtedly due to an increase in the amount of transmitter released, as demonstrated in radiochemical experiments (Fig. 2); samples treated with oxythiamine released more ACh than

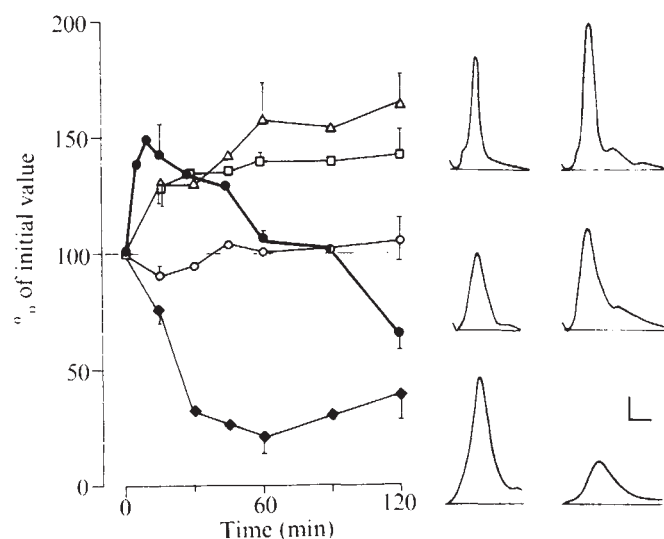


Fig. 1 Effects of external thiamine, oxythiamine and adenosine on the electrical discharge. Pieces of tissue containing one to three prisms were excised from the electric organ and their response to single shocks were tested at long time intervals. The prisms are made of 400–500 superposed electroplaques and, when submitted to nerve or field stimulation, they generate an electrical discharge which is the sum of a large number of synchronised postjunctional potentials^{5,7,8}. When incubated in normal physiological medium, the tissue produced, for several hours, constant responses to single stimulations. The right hand upper tracings in the figure show discharges recorded before and after a 60-min incubation in 5×10^{-3} M thiamine (calibration: 0.2 V, 2 ms). Middle records: effect of 10^{-4} M oxythiamine for 45 min (0.5 V, 2 ms). The effect of oxythiamine was related to its concentrations in the 10^{-5} – 10^{-3} M range. Lower records: 5×10^{-3} M adenosine for 60 min (0.2 V, 2 ms). In the graph, the surfaces of the discharge are expressed as percentage of their initial values and plotted as a function of time; s.e. is indicated only for crucial points. Δ , 5×10^{-3} M thiamine; $\square$, 10^{-4} M oxythiamine; $\circ$, controls; $\bullet$, 10^{-2} M thiamine; $\blacklozenge$, 5×10^{-3} M adenosine.

the controls with the most striking difference observed during the first 2 min of continual stimulation.

The same methods were then used to study adenosine, which has been reported to reduce “quantal release” at the neuromuscular junction¹⁰. The effect of this compound, unlike that of oxythiamine, was to reduce the electrical discharge and decrease the quantity of ACh released (Figs 1 and 2). Addition of 10^{-3} M adenosine to the medium could reverse the effect of 10^{-4} M oxythiamine in 15 min.

Experiments in which 10^{-3} M TPP was added to the medium, showed that this ester had no effect on synaptic transmission, probably because of its inability to penetrate cell membranes⁴. The effects of thiamine itself were a function of concentration and time (Fig. 1). At a concentration of 10^{-2} M, thiamine first caused an increase in

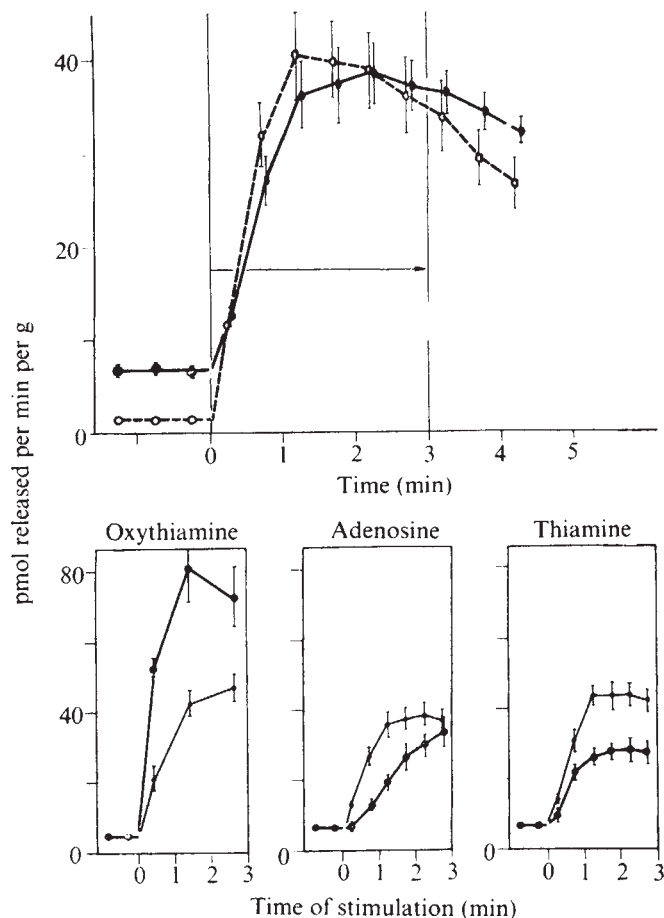


Fig. 2 Release of radioactive ACh from pieces of the *Torpedo* electric organ. The tissue was incubated for 4 h in physiological medium in the presence of ^{14}C -acetate (—, 132 d.p.m. μmole^{-1}) and ^3H -choline (---, 1,320 d.p.m. μmol^{-1}) at the same concentrations ($8.4 \mu\text{M}$). In this tissue, the two precursors are incorporated into ACh^{14,15}. The fragments were washed overnight and then submitted to continual stimulation at 10 per s for 3 min during superfusion at 1 ml min^{-1} . ^{14}C and ^3H radioactivities were counted in the superfusing solution and provided a sensitive and direct measurement of the ACh released during stimulation. At rest, the level of radioactive choline was constantly less than that of acetate, but, on stimulation, they increased in a ratio of 1:1. Lower graphs, Effects of 10^{-4} M, oxythiamine, 10^{-3} M adenosine and 5×10^{-3} M thiamine after 1 h of incubation. Only the ^{14}C values are shown here because they were not significantly different from the ^3H values. The fine lines indicate controls. In the oxythiamine experiment, ^3H -acetate and ^{14}C -choline were used. Adenosine (5×10^{-3} M) had the same effects as 10^{-3} M. In the presence of adenosine, the electrical discharge started at a lower level than in controls, but it increased during stimulation, an effect already described for ATP in this tissue¹⁶. Thiamine (10^{-3} M) increased slightly, but not significantly, the amount of ACh released.

Table 1 Comparison between the phosphorylation of thiamine and the level of “free” ACh in the electrogenic tissue of *Torpedo* in different conditions

Extraction after	Thiamine (%)	TMP (%)	TPP (%)	“Free” ACh (nmol g ⁻¹)
Incubation in the physiological medium (11)	28.6 ± 2.5	28.6 ± 5.5	42.8 ± 5.2	296 ± 29
Homogenisation in the physiological medium (8)	46.1 ± 6	28 ± 8	26 ± 5.7	0
Incubation in the medium with 5.3 mM Mg ²⁺ and 0.01 mM Ca ²⁺ (4)	62 ± 15	4 ± 4	34 ± 13	58 ± 27

Pieces of electric organ were excised under Tricain anaesthesia and placed in a physiological medium suitable for elasmobranch fish⁶. Thiamine and its phosphoric esters were extracted with 5% trichloroacetic acid, separated by high voltage electrophoresis and their concentration was determined by the fluorimetric procedure of Itokawa and Cooper¹². ACh was measured in the same extracts by a radiochemical assay¹³. The values of thiamine, TMP and TPP are expressed as percentage ($\pm$ s.e.) of the total content of thiamine plus esters; this total was $458 \pm 92 \text{ nmol g}^{-1}$ wet weight and did not show significant variations between the three conditions. “Free” ACh was determined as the difference between the total ACh ($708 \pm 64 \text{ nmol g}^{-1}$) and the ACh remaining after homogenisation of the tissue⁴. Thus “free” ACh was equal to zero by definition in the second condition. Numbers of experiments are given in parentheses.

the electrical discharge but, in a second phase, the discharge decreased considerably, the effects of thiamine thus becoming similar to that of adenosine. At a lower concentration (5×10^{-3} M) of thiamine, amplification of the discharge lasted longer but after 1 h transmitter release had already diminished (Figs 1 and 2). This apparent discrepancy is due to the thickness (4–6 mm) of the pieces of tissue, through which thiamine requires time to diffuse⁶, since it is from the surface of the samples that the transmitter is collected, but it is chiefly at the core of the tissue where the discharge is generated. The effect of thiamine in the first phase differed from that of oxythiamine in that it was essentially the amplitude of the electrical discharge that was increased rather than the duration. This effect was transient, however, for in a second phase, thiamine, particularly at a high concentration, caused a sharp reduction of the discharge, related to the decrease in the quantity of ACh released.

The rather unexpected electrophysiological changes observed in this study are unlikely to be due to modifications of the rate of ACh hydrolysis by acetylcholinesterase, for the inhibitors of this enzyme are known to alter nerve-electroplaque transmission in a different way⁵. This was confirmed in biochemical tests which revealed that oxythiamine and adenosine, at the given concentrations, had no effect on the activity of cholinesterase extracted from the electric organ. Thiamine was also found not to inhibit the rate of hydrolysis except at a concentration higher than that of the ACh used as substrate. This might be due to the capacity of thiamine to form complexes with ACh, which have been found to be increasingly stable as the proportion of thiamine is increased¹¹.

It can be concluded from the observations of endogenous thiamine and ACh release, that the presynaptic nerve endings are the most probable site of thiamine involvement in synaptic transmission. It seems, moreover, that thiamine might even be directly implicated in transmitter release. A possible mechanism to explain its role would be as follows: thiamine might, in some way, bind to a fraction of "free" ACh, the pool of transmitter available for release⁶⁻⁸. This fraction, under the influence of Ca^{2+} , would then be released in the synaptic cleft, perhaps in a quantal manner. The release would be accompanied by a transfer of phosphate either to or from the thiamine and might occur at the presynaptic membrane, possibly with the involvement of synaptic vesicles. External oxythiamine as well as thiamine in the first phase, would cause an increase in the fraction of transmitter loaded and released, whereas adenosine or an excess of thiamine would cause a decrease.

Further investigations are needed to establish the validity of this theory as well as to determine whether thiamine is also involved in the release of other transmitter substances. Our results, however, suggest that the requirement of the vitamin B_1 by the nervous system can be better explained by its involvement in synaptic transmission rather than in axonal conduction.

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An inhibitory tripeptide from cat spinal cord

UNTIL quite recently, the possibility that naturally occurring peptides, with a few exceptions, such as substance P, might function to modify the excitability of neurones in the central nervous system (CNS) was considered rather remote. There is now increasing evidence to suggest that peptides have a greater range of actions and are more widely distributed than previously thought¹⁻⁶. In a search for physiologically active peptides in the CNS we have isolated a tripeptide from the dorsal columns of cat spinal cord, which has been partially characterised as His-(Gly, Lys). We have synthesised the two possible forms and tested their activity by iontophoretic application to single neurones in the spinal cord, the medial medulla oblongata and the dorsal column nuclei.

To extract peptides from central nervous tissue we used a technique which minimises degradative processes⁷. Lumbar segments L2–L5 of the spinal cord were exposed in barbiturate-anaesthetised cats and frozen *in situ* with liquid nitrogen. The frozen tissue was excised and allowed to thaw to 4 °C, when the dorsal columns were dissected out and homogenised in acetone: 1 M HCl (100:3 v/v) at –20 °C. The homogenate was stirred at 4 °C for 24 h. After centrifugation, the supernatant was lyophilised, suspended in a small volume of isopropanol-water (10:90 v/v), and applied to a Sephadex G-15 column. Peptides were detected in the column effluents by their absorption at 206 nm. One of the G-15 column fractions was run on a DEAE-Sephadex column to separate the peptides further and a fraction from this column was analysed by thin-layer chromatography.

Two separate methods were used to isolate individual peptides from this fraction. Dansylated peptides were chromatographed in two dimensions on polyamide plates; non-dansylated peptides were chromatographed on silica gel plates and located using fluorescamine. Fourteen components were detectable and one of them (that with the strongest fluorescence) had the R_f values shown in Table 1. This component, a discrete spot, was scraped from the polyamide plate. After hydrolysis, it was rerun on a second polyamide plate, when only dansyl-histidine could be detected, indicating that the N-terminal amino acid was histidine. The remainder of this second plate was eluted and dansylation of the concentrated eluate yielded only glycine and lysine on further chromatography. Since the elution time on Sephadex G-15 suggested a tripeptide structure, we concluded that the peptide was His-(Gly, Lys).

The two possible tripeptides His-Gly-Lys and His-Lys-Gly

Table 1 R_f values of the tripeptide His-(Gly, Lys) from cat spinal cord

	First dimension Solvent 1	Second dimension Solvent 2
Non-dansylated peptide (silica gel F254 plate)	0	47
Dansylated peptide (polyamide-6 plate)	Solvent 3 85	Solvent 4 5

The solvents were as follows: 1, butanol-acetic acid-water (4:1:1 by volume); 2, pyridine-acetic acid-water (10:6:3 by volume); 3, water-formic acid (200:3 v/v); 4, benzene-acetic acid (9:1 v/v).

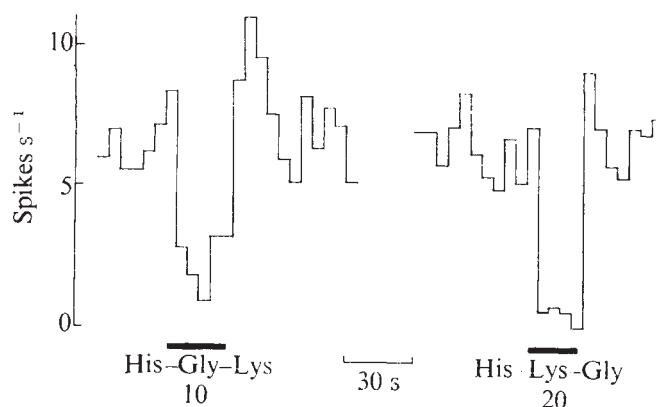


Fig. 1 The effects of iontophoretically applied His-Gly-Lys and His-Lys-Gly on the firing rate, induced by a continuous current of 4 nA of L-glutamate, of a neurone in the dorsal horn of the lumbar spinal cord. Na^+ (20 nA) had no effect. Applications are indicated by the horizontal bars; currents are shown in nA. Ordinate: Mean firing rate in impulses s^{-1} in successive 5-s epochs. Solutions of the peptides were made up to 0.05 M, pH 4.1–4.5.

were synthesised by conventional methods by two different routes. Both utilised mixed anhydrides or azides for coupling but differed in the type of protective groups used. In one set of syntheses, the carboxyl terminus was protected with benzyl, the sidechain of Lys and the α -amino group of His with benzyloxycarbonyl. The alternative scheme utilised *tert*-butyl for the carboxyl terminus and *tert*-butoxycarbonyl for the Lys side-chain and His α -amino group. After removal of the protective groups by hydrogenolysis or protonolysis, respectively, the free peptides were purified by ion-exchange chromatography on carboxymethyl cellulose to yield homogeneous materials as shown by thin-layer chromatography and electrophoresis in several systems. The products of all four syntheses contained the correct ratio of amino acids after both acid hydrolysis and digestion with aminopeptidase M, indicating absence of racemisation during synthesis. H-His-Lys-Gly-OH triacetate had $[\alpha]_D^{26} = -12.3^\circ$ ($C = 1$, water) whereas H-His-Gly-Lys-OH triacetate had $[\alpha]_D^{26} = +5.6^\circ$ ($C = 0.8$, water).

The two synthetic peptides and the endogenous peptide had identical R_f values in the thin-layer chromatographic systems used and the same elution time on the Sephadex G-15 column. Consequently, the amino acid sequence of the natural peptide could not be assigned by direct chromatographic comparison with synthetic samples and will have to await the isolation of further material. Nevertheless, the characterisation of the isolated peptide as His-(Gly, Lys) is supported by the identical chromatographic behaviour of the synthetic and naturally occurring compounds.

Both synthetic tripeptides were applied by iontophoresis to single neurones in three regions of the CNS in unanaesthetised, decerebrate, decerebellate cats (mid-collicular decerebration was performed under halothane anaesthesia). Since the natural peptide was extracted from the dorsal columns, the responses of neurones in the dorsal column nuclei were examined first, and this was followed by application to cells in the spinal cord and the medial brainstem. The peptides were tested for effects on spontaneous activity or, when this was low or absent, on activity induced by the continuous application of the excitant L-glutamate from another barrel of the micropipette.

The dorsal column nuclei were located from surface landmarks and neurones were identified by their localised ipsilateral receptive fields in response to hair movement or joint movement or touch. Of 24 cells tested with His-Gly-Lys, 21 (12 in the cuneate nucleus and nine in the gracile nucleus) were unaffected by currents up to 150 nA, whereas three neurones in the gracile nucleus were inhibited at currents from 50–100 nA. His-Lys-Gly was tested on 15 cells; one in the gracile nucleus was

inhibited and the remainder in both dorsal column nuclei showed no response.

In the spinal cord (L3–L5), the peptides affected a much larger proportion of neurones. When applied with currents of 10–50 nA His-Gly-Lys inhibited (Fig. 1) 17 (40%) of 42 neurones, the remainder being unaffected. The positions of four cells inhibited by His-Gly-Lys were marked with pontamine blue, and all of these were in the dorsal horn; three seemed to be in lamina V and one was at the border of laminae VI and VII. His-Lys-Gly was tested on six cells, two of which were inhibited.

To test the peptides on cells in the medial brainstem, micropipettes were inserted through the floor of the IVth ventricle between 1.0 and 4.8 mm rostral to the obex and 1.0–1.5 mm lateral to the midline. As in the spinal cord, both peptides had depressant effects on some cells. His-Gly-Lys inhibited 30 (52%) of 58 units and His-Lys-Gly 11 (37%) of 30 units. Some of these neurones were identified as reticular by their response to somatosensory stimulation over a large area of body surface.

In all three regions of the CNS the reduction in firing rate was rapid in onset, beginning within the first second of the ejection period and reaching a plateau within 15–20 s (Fig. 1). At the end of the application the firing rate returned rapidly to its previous level. The degree of inhibition was related to the strength of iontophoretic current, and clear responses were sometimes seen with currents as low as 10 nA. No excitatory effects were seen. In most cases when similar currents were used to eject the two compounds, His-Gly-Lys produced a greater degree of inhibition than His-Lys-Gly. In all, His-Gly-Lys inhibited 50 of the 124 units examined (40%) whereas His-Lys-Gly inhibited only 14 of 51 units (27%). The former thus seems to be more potent than the latter, although a more accurate comparison of potencies cannot be made without knowledge of the relative efflux rates of these substances from each micropipette, in relation to the current applied⁸.

The tripeptide His-(Gly, Lys) thus has inhibitory effects on some neurones in the dorsal horn of the spinal cord and in the medial brainstem, including the reticular formation. Although it was found in the lumbar dorsal columns it has little effect in the dorsal column nuclei, and since the ascending fibres to these nuclei are excitatory it is unlikely to be concerned in transmission in this system. The dorsal columns do, however contain other fibres, such as propriospinal and reticulospinal⁹, and the peptide may be associated with intersegmental fibres or descending fibres terminating in the dorsal horn. The occurrence of the tripeptide His-(Gly, Lys) in the spinal cord and its actions on selected neurones in the dorsal horn suggest that it may have a functional role in relation to neuronal inhibition in the spinal cord.

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Di- and trimethylated congeners of 7-methylguanine in Sindbis virus mRNA

In the past two years it has become clear that most animal messenger RNA—both cellular¹⁻⁶ and viral⁷⁻¹¹—is “capped” by 7-methylguanosine (m^7G) residues present in inverted nucleotide linkage at 5' termini of the general form $m^7G(5')pppNm$. At least for certain mRNAs the capping m^7G has an important role in mRNA-ribosome binding and thus in initiation of protein synthesis¹²⁻¹⁵. We now report novel forms of capped 5' termini on the main mRNA, '26S' RNA, coded for by the animal alphavirus, Sindbis. These termini contain, in addition to the usual m^7G , its hypermodified congeners $m_2^{2,7}G$ and $m_3^{2,2,7}G$. This last compound has been previously described only in small nuclear (sn) RNAs of unknown function^{16,17}, whereas $m_2^{2,7}G$ has not been described previously.

Caps were obtained from methyl-³H-methionine-labelled 26S RNA, using T2-ribonuclease followed by DEAE-cellulose chromatography¹⁸. The recovered oligonucleotides were degraded to ribosides using bacterial alkaline phosphatase and snake venom phosphodiesterase¹; only the extreme 5'-terminal ribosides of such oligonucleotides were labelled, since 26S RNA lacks penultimate Nm residues¹⁸. Preliminary analyses indicated the presence of three methyl-labelled species: m^7G plus two other ribosides whose properties suggested that they were also 7-methylated derivatives of guanosine, possibly $m_2^{2,7}G$ and $m_3^{2,2,7}G$ (data not shown). Accordingly, we prepared marker compounds by reacting m^2G and m_2^2G with dimethyl sulphate in conditions known to result in

preferential methylation at the 7 position^{16,19}. Evidence confirming that the products were indeed the expected $m_2^{2,7}G$ and $m_3^{2,2,7}G$ is provided by mass spectra of their free bases (Fig. 1). The most intense peak in the spectrum of both compounds is that of the molecular ion. The assignment of major peaks in Fig. 1a is: m/e 179, the molecular ion M ; m/e 123, 95, 68, arising from the successive loss of cyanamide fragment (CH_3)HNCN, CO and HCN; m/e 150, possibly by the loss of CH_3N . The assignment of major peaks in Fig. 1b is: m/e 193, the molecular ion M ; m/e 123, 95, 68, resulting from sequential expulsion of cyanamide fragment (CH_3)₂NCN, CO, and HCN; m/e 178, resulting from the loss of one methyl group; and m/e 164, possibly arising from expulsion of CH_3N . The loss of the methylamino group resembles that of N^6 -methylated and N^6 -dimethylated derivatives of adenine. The initial expulsion of a neutral cyanamide fragment with sequential elimination of CO and HCN is highly characteristic of methylated guanines²⁰. Below m/e 120, the mass spectra of both $m_2^{2,7}Gua$ and $m_3^{2,2,7}Gua$ are qualitatively similar to that of m^7Gua (refs 20 and 21). For precise comparison with ³H-labelled samples from 26S RNA we also prepared ¹⁴C-methyl-labelled $m_2^{2,7}G$ and $m_3^{2,2,7}G$ in the same manner.

Figure 2 shows a phosphocellulose elution pattern obtained for the methylated ribosides of the 26S RNA cap, following the procedure used in the original isolation of $m_3^{2,2,7}G$ (ref. 16). The two unknown peaks (I and II in Fig. 2) correspond precisely to the markers $m_3^{2,2,7}G$ and $m_2^{2,7}G$, respectively. The third major ³H peak III corresponds, as expected, to m^7G . The radioactivity in the void volume (V) probably represents partial degradation products of the relatively labile 7-methyl-purine ribosides^{16,23,24}.

Table 1 summarises the behaviour of m^7G , $m_2^{2,7}G$, and $m_3^{2,2,7}G$, and their bases, in four other systems. In all cases the ³H from peaks I and II (Fig. 1), or the corresponding bases released by acid hydrolysis, ran with $m_3^{2,2,7}G$ and $m_2^{2,7}G$, respectively, or their corresponding bases (these latter were the 'unidentified' bases in our initial report¹⁸ on acid hydrolysates of 26S RNA). We conclude that ribosides I and II are indeed $m_3^{2,2,7}G$ and $m_2^{2,7}G$, respectively.

The $m_3^{2,2,7}G$ residues of snRNAs U-1 and U-2 have been shown to occur as caps on the 5'-terminal sequence $m_3^{2,2,7}G(5')pppAmUm \dots$ (refs 25 and 26), which resembles that of 26S RNA, $mG(5')pppApUp \dots$ (ref. 27 and Dubin *et al.*, unpublished). Conceivably these two forms of RNA, although apparently grossly different in biological and chemical properties, are substrates for the same or related methylases. Another possibility suggested by the presence of similar caps

Fig. 1 Mass spectra of $m_2^{2,7}Gua$ (a) and $m_3^{2,2,7}Gua$ (b). Dried samples of ribosides prepared as described in the text were dissolved in 1 N HCl, heated for 45 min at 100 °C, adjusted to pH 8.5 with concentrated NH_4OH , and lyophilised to dryness. The mass spectral analysis was carried out at 70 eV using a direct inlet system at a temperature of 270 °C.

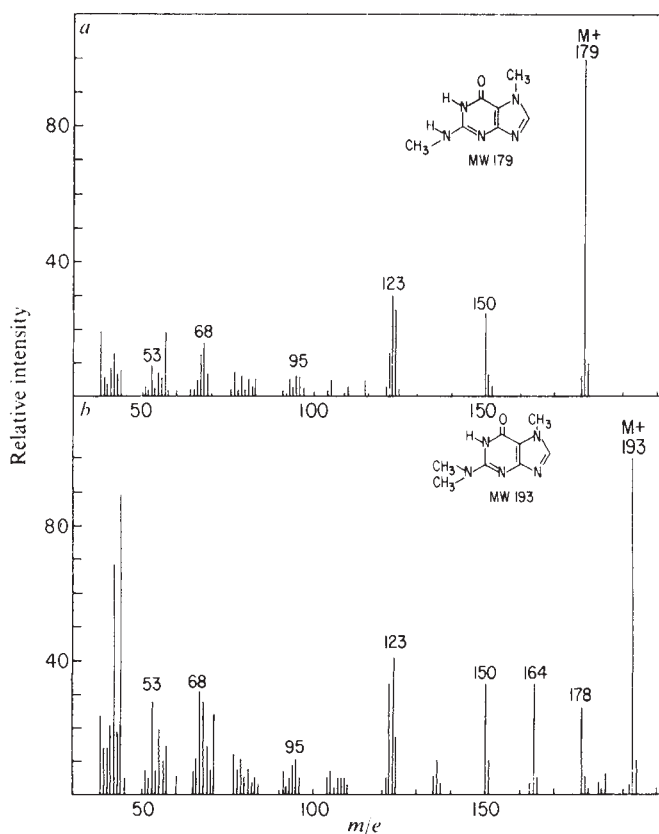


Table 1 Chromatography and electrophoresis of 7-methylated guanines and guanosines

	Mobility or R_f relative to adenine Electrophoresis, pH 3.5	Solvent system		
		A	B	C
m^7G	1.18	1.25	0.15	0.54
$m_2^{2,7}G$	1.18	2.06	0.45	0.89
$m_3^{2,2,7}G$	1.18	1.85	0.89	1.33
m^7Gua	0.50	0.85	0.57	—
$m_2^{2,7}Gua$	0.55	1.93	1.06	—
$m_3^{2,2,7}Gua$	0.73	1.70	1.12	—

Electrophoresis and chromatography were carried out as described previously^{1,18}. Chromatographic systems were: A, Whatman 3MM paper developed in isopropanol-11.6 N HCl- H_2O (60:17.6:22.4, by volume); B, Whatman 3MM paper developed in n -BuOH- H_2O (86:14, by volume in NH_3 atmosphere); C, thin-layer cellulose sheets developed in ethyl acetate-isopropanol-7.5 M NH_4OH - n -BuOH (3:2:2:1, by volume). For convenience we have expressed values relative to adenine, which is readily available and which moves well in all systems. R_f for adenine in systems A, B and C were about 0.29, 0.25 and 0.40; it ran about 20 cm towards the cathode in 3 h at 3,000 V. Abbreviations follow conventional usage (for example, m^7Gua , 7-methylguanine; m^7G , 7-methylguanosine).

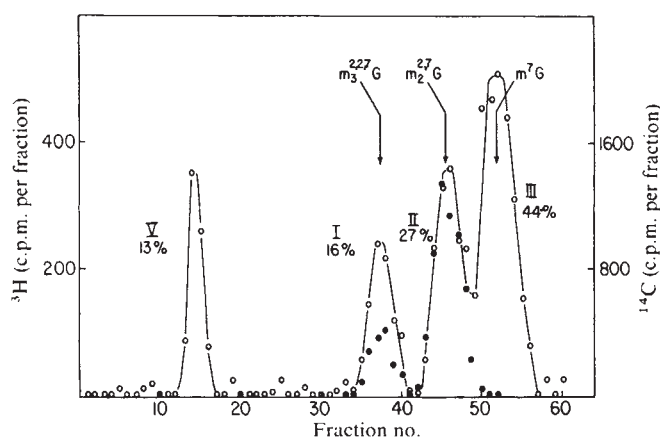


Fig. 2 Phosphocellulose pattern of methylated ribosides from the Sindbis virus 26S RNA cap. The 5'-terminal oligonucleotide of methyl- ^3H -methionine-labelled Sindbis 26S RNA was prepared from infected chick cells and digested to its constituent ribosides as described previously^{1,18,22}. The digest was chromatographed on a phosphocellulose column using a linear gradient of 0.01–0.3 M ammonium acetate¹⁶. As markers we added ^{14}C -methyl-labelled $m_3^{2,7}\text{G}$ and $m_3^{2,2,7}\text{G}$ and unlabelled $m^7\text{G}$. Arrows indicate the midpoints of the 260-nm absorbancy peaks of the markers, and the arabic numeral next to each peak indicates the percentage of the endgroup ^3H present in that peak. $\circ$, ^3H ; $\bullet$, ^{14}C .

on sn and 26S RNAs is that our hypermethylated congeners—or at least $m_3^{2,2,7}\text{G}$ —are not in 26S RNA proper but rather occur in snRNA–26S RNA complexes. We believe this is unlikely, since 26S RNA lacks the Nm groups that are abundant in snRNA; our infected cells were labelled in the presence of a level of actinomycin ($4\text{ }\mu\text{g ml}^{-1}$, ref. 18) that should substantially suppress cellular RNA synthesis; and recent studies (Dubin *et al.*, unpublished) indicate the presence of 1 mol $m^7\text{G}$ plus congeners per 26S RNA molecule.

There is at present no evidence for a biological role of the extra methyl groups of the 26S RNA capping residues. We are currently attempting to explore one obvious possibility—that they enhance affinity to ribosomes—by comparing poly-some-bound and free 26S RNA.

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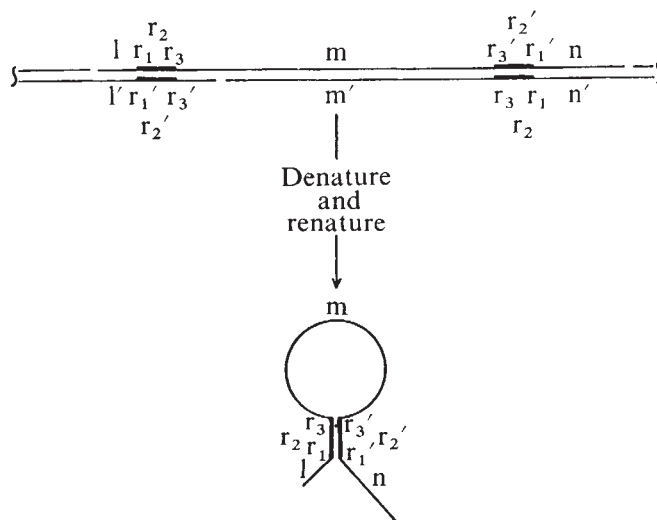
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Distribution of inverted IS-length sequences in the *E. coli* K12 genome

THE genomes of *E. coli* and some of its plasmids contain a class of elements called insertion sequences (IS)^{1–5}. These elements exhibit various properties associated with modulation and alteration of gene expression. IS1 (length 800 nucleotide pairs^{2,3}) apparently plays a role in the amplification of resistance determinants in drug resistance factors^{5,6} and also causes polar insertion mutations^{2,3}. IS2 (length 1,400 nucleotide pairs²) behaves as a controlling element, which can turn gene expression on or off⁷. IS3 (length 1,400 nucleotide pairs³) has been associated with strong polar mutations, and is a constituent of a transposable drug resistance determinant⁸. IS2 and IS3 have also been shown to specify integration loci for the fertility plasmid F^{4,9,10}. Hybridisation data have allowed estimation of the number of IS1 and IS2 elements in the *E. coli* genome¹, but except for the region around *lac* and *purE*, little is known about their natural location in the genome.

While investigating the sequence organisation of three Hfr strains of *E. coli*, we encountered a number of intrastrand hybrid structures which provide information on the distribution of inverted sequences within the *E. coli* K12 genome. Figure 1

Fig. 1 Formation of intrastrand DNA hybrid structures. The upper portion of the figure depicts a slightly nicked DNA duplex. The letters each denote a block of nucleotides; the sequences complementary to each block are indicated by the corresponding primed letter. The sequence $r_1r_2r_3$ is repeated on the DNA molecule in inverted order. Random nicking of DNA duplex (or shear breakage of double-strand or single-strand DNA) produces some strands which have both the sequences $r_1r_2r_3$ and the complementary sequences $r_3'r_2'r_1'$, as indicated for the upper strand. After denaturation and the onset of annealing, the inverted repetitions on such strands can rapidly form intrastrand hybrids, since $r_1r_2r_3$ and $r_3'r_2'r_1'$ are physically linked. Other strands, like the lower one, contain breaks between the inverted sequences, and in such cases, the inverted repetitions will renature slowly since bimolecular processes are required. The intrastrand hybrid structure formed from the upper strand is shown at the bottom. The length of the duplex stem corresponds to the length of the inverted repetition, and the length of the single-strand loop indicates the spacing between inverted repeats.



shows that if a pair of sequences is present as an inverted repetition, denaturation and subsequent reannealing of the DNA may result in an intrastrand hybrid structure. The repeated sequences form a duplex stem whose length corresponds to the length of the repeated sequence. The DNA separating the inverted sequences appears as a single strand loop whose length corresponds to the distance separating the inverted repetitions. Directly repeated sequences will not form intrastrand duplexes. Figures 2 and 3 are electron micrographs of two different intrastrand hybrid structures obtained from the DNA of strain P804. Similar structures were seen with DNA extracted from the strains P4X and P3. In both figures, the length of the duplex stem is approximately 1,300 nucleotide pairs (1.3 kilobases, or kb).

Three size classes of single-strand loops were detected (Table 1). There were only two representatives of the largest size class. Intrastrand hybrid structures with larger loops were less abundant than structures with smaller loops. Since renaturation times were long, the difference in abundance of these intrastrand structures is unlikely to be the result of kinetic effects. Because DNA strand lengths for a typical preparation were 120 ± 60 kb, inverted repetitions separated by more than 180 kb could form loop structures only rarely. Inverted repetitions located far from each other are more likely to be separated by strand breaks between the inverted sequences than are closely linked inverted repetitions, and therefore larger loop sizes will appear less frequently than smaller ones, even though the more widely spaced inverted sequences may appear in the genome at the same frequency as the less widely spaced ones. This relative enrichment for smaller loop sizes is reflected in Table 1.

These experiments do not identify the DNA contained in the duplex stems; however, the stem sizes are approximately

Fig. 2 Intrastrand hybrid structure from *E. coli* K12 strain P804. Folded chromosomes were isolated by using modifications of the methods of Worcel and Burgi^{10,15}. Bacterial DNA was denatured by incubation in 0.33 M NaOH for 5 min at 37 °C, and then for 10 min at room temperature. The sample was neutralised and dialysed at room temperature for 2–3 h against 70% formamide, 0.25 M NaCl, 0.10 M Tris, 0.01 M EDTA, pH 8.5, for the reannealing step. The concentration of the bacterial DNA was approximately $1.5 \mu\text{g ml}^{-1}$. Renaturation was terminated by dialysis at 4 °C against 50% formamide, 0.10 M Tris, 0.01 M EDTA pH 8.5. DNA was mounted from 55% formamide by the basic protein film technique as previously described¹⁶. The arrow in the micrograph points to the duplex stem, whose length is 1.2 kb. The single-strand loop has a length of 29 kb. The scale bar indicates the distance corresponding to 1 kb (1,000 nucleotides of single-strand DNA or 1,000 nucleotide pairs of double-strand DNA). Circular reference molecules are from ϕX174 .

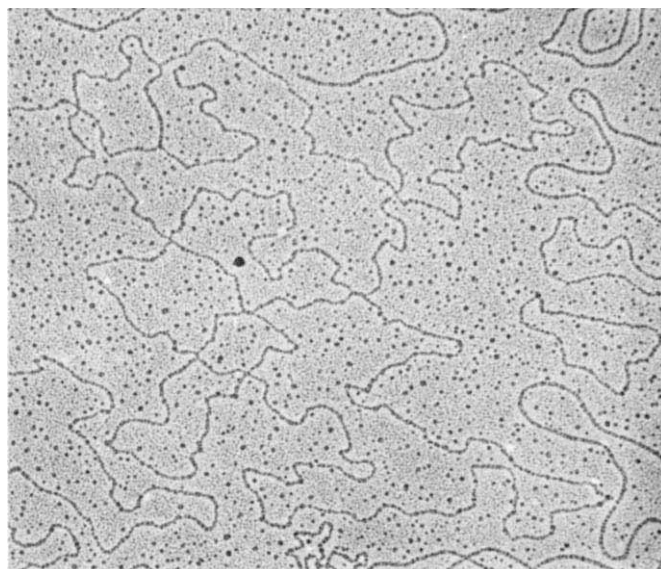
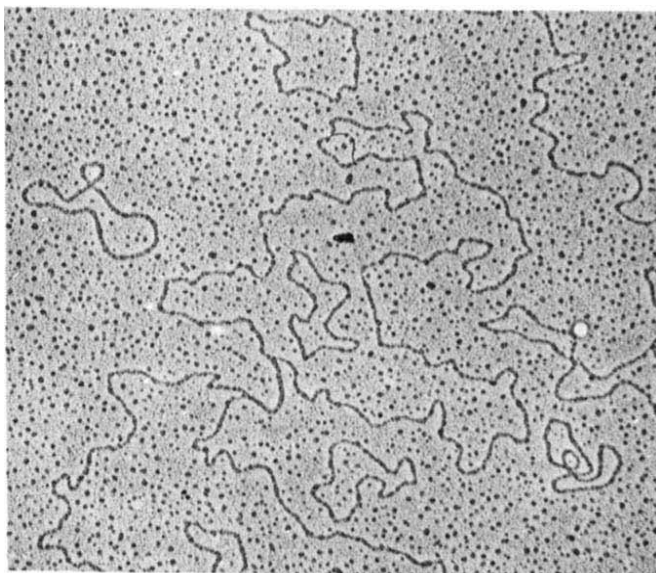


Fig. 3 Intrastrand hybrid structure from *E. coli* K12 strain P804. The stem (indicated by the arrow) has a length of 1.3 kb, and the loop has a length of 69 kb.

the same size as both IS2 and IS3. The duplex stem length associated with the 69-kb loops is 1.3 ± 0.1 kb, while the stem length for the 29-kb loop structure is 1.2 ± 0.2 kb. Histograms of stem lengths are shown in Fig. 4. The histogram for hybrid regions from the 29-kb structures (Fig. 4a) displays a shoulder corresponding to shortened stem lengths. Such shortening could result from partial melting of a partially mismatched structure, or it could indicate that the stems of the 29-kb loops are shorter and therefore different from the stems of the 69-kb loops.

Since the 0.8-kb IS1 element is known to be present in the *E. coli* genome¹, there is the possibility that intrastrand hybrid structures involving IS1 might form. Three intrastrand structures with duplex stems shorter than 1 kb (and distinct from the structures with 29-kb loops) were detected, but each displayed a different loop size. We would have detected any structure classes having hybrid portions in the 0.75–1.00-kb size range had they been present at frequencies comparable with those of the other structures. As Fig. 4a shows, a few examples of the 29-kb structure with duplex regions in this size range were seen, which confirms that our specimen scanning procedures were sensitive to this size range. We conclude that

Table 1 Distances separating 1.3-kb inverted repetitions in three strains of *E. coli* K12

Strain	Loop sizes (kb)		
	I	II	III
P3	29.4 ± 2.6 (11)	67.4 ± 3.8 (9)	184.7^* (1)
P4X	29.5 ± 1.9 (6)	73.1 ± 4.1 (4)	—
P804	29.3 ± 1.4 (10)	69.6 ± 2.6 (5)	173.1 (1)
Average	29.4 ± 2.0	69.3 ± 4.1	

Intrastrand hybrid structures were detected by scanning denatured and reannealed preparations of *E. coli* DNA in the electron microscope. Sizes were calculated by using ϕX174 DNA mounted on the same grid as a size reference (taken to be 5.25 kb). The numbers of structures measured are given in parentheses.

*This structure contained two hybrid regions whose sizes and relative separation suggested the participation of sequences from the integrated F plasmid.

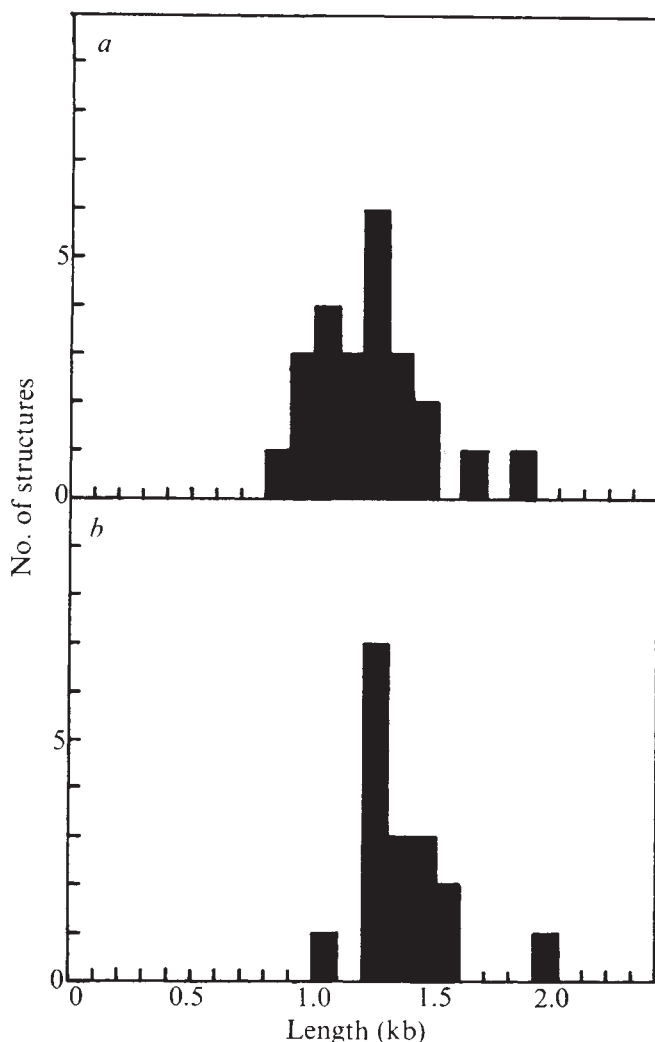


Fig. 4 Histograms of lengths of duplex hybrid regions for the major loop size classes: *a*, hybrid lengths associated with the 29.4-kb loop; *b*, hybrid lengths associated with the 69.3-kb loop.

there are probably few IS1 length inverted repetitions relatively close together in the strains which we have examined; however, we cannot rule out the possibility that our reannealing or mounting conditions are somehow unfavourable for formation or retention of such structures. It is important to note that inverted repetitions longer than 1.3 kb would have been detected if they were sufficiently close. No such structures were observed.

In addition to the inverted repeat structures described, more complicated examples containing two hybrid regions were seen, but these are attributable to participation of IS2 and IS3 elements present on the integrated F factor in these Hfr strains. It should be emphasised that the organisation of the integrated F in these three strains is known¹⁰ (our unpublished results), as are the structures of the added F and F-prime heteroduplex reference molecules which were present during these experiments. The structures which appear in Figs 2 and 3 are not characteristic of the F or F-prime reference DNA which was also present. The main size classes seen in strain P3 are found also in P4X and P804, even though the site of the integrated F factor in P3 is some 240 kb removed from the F integration site in P4X and P804. This provides further evidence that these inverted repeat structures are not dependent on the Hfr character of these strains.

The data presented here provide information on the distribution of IS2 and IS3 size inverted repetitions in the *E. coli*

K12 genome. Previous heteroduplex mapping of the F-prime F13, which carries the *lac-purE* region from *E. coli*, identified three IS3 elements and one IS2 in the region⁹. If these elements are normal constituents of *E. coli* K12, then the 69-kb loop is predictable from separation distances and orientations of IS3 elements in the *lac-purE* region. The location of the DNA contained in the smallest loop is unknown.

We can conclude that elements similar in structure and size to the *tet*^R transposon⁸ and the *kan-2* elements¹¹, which have flanking inverted repetitions of 1.4 kb, are absent in the strains which we have examined. The loop sizes which are seen in intrastrand hybrid structures containing these elements are in the 5–10-kb size range, and we would surely have detected them. Elements structurally similar to the TEM β -lactamase transposon TnA¹² would not have been detected in these experiments, since the short stems characteristic of their intrastrand hybrid structures would have looked like overlaps.

The Hfr strains P4X and P3 are closely related¹³, and they exhibit similar loop sizes. P804 is less closely related to P3 than is P4X¹³, yet the size classes correspond to those of P3 and P4X. This again suggests that these inverted elements are relatively stable components of the *E. coli* genome, as has been suggested for IS3¹⁴.

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Cleavage map of colicin E1 plasmid

COLICIN E1 plasmid (colE1) is a closed circular DNA molecule with a molecular weight of 4.2×10^6 (ref. 1). ColE1 DNA has extensively been used as a molecular vehicle for cloning and amplification of DNA in genetic engineering^{2,3}. In order to expand such investigations, it is useful to make a cleavage map ordering colE1 DNA pieces produced by bacterial restriction endonucleases. *Escherichia coli* RI restriction endonuclease (R·EcoRI) has been shown to cleave colE1 DNA at a single unique site^{4–6}. We have now constructed a physical map with the EcoRI site as a reference point using two restriction endonucleases from *Haemophilus aegyptius* (R·HaeII and R·HaeIII). The map of the colE1 moiety involved in a hybrid plasmid pML21 (ref. 7) carrying about a half of the colE1 genome was also determined. Restriction endonucleases and fragments pro-

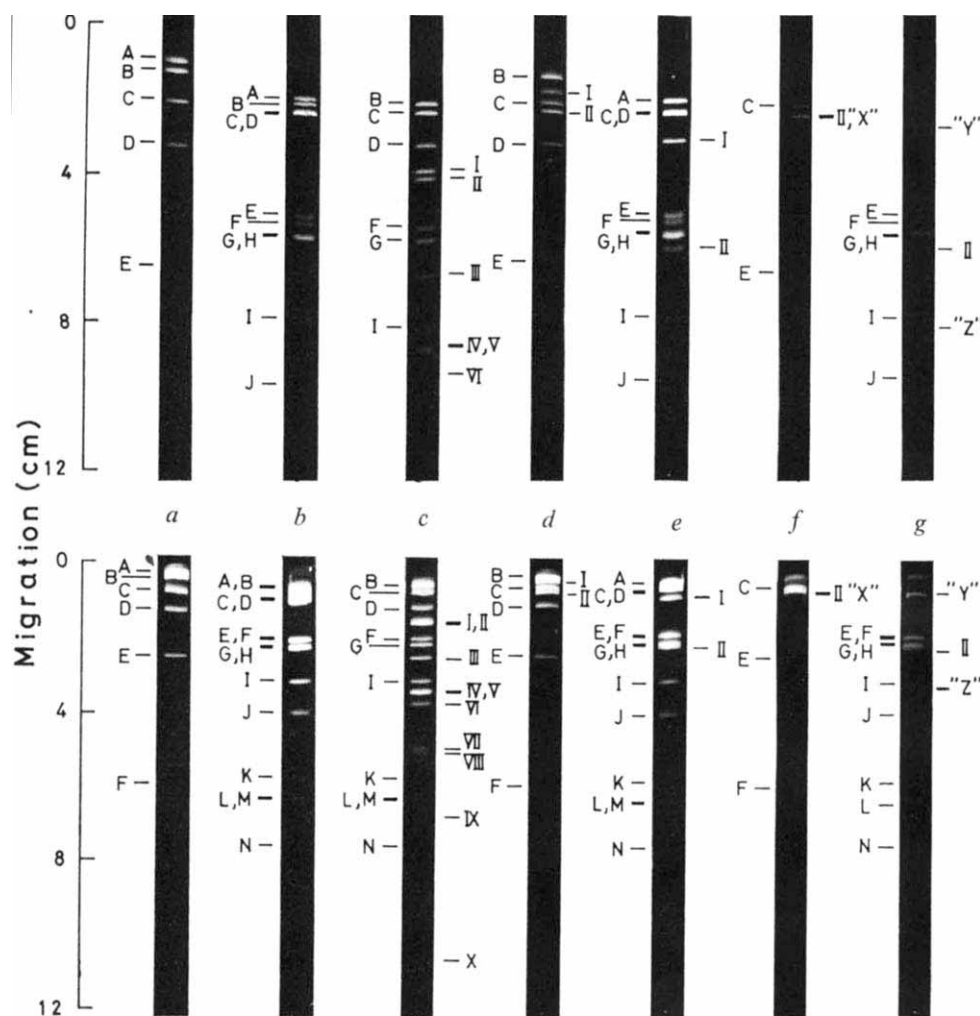


Fig. 1 Gel electrophoretic patterns of digests of colE1 DNA and the mini-colE1 segment of pML21 with restriction endonucleases. ColE1 DNA or the mini-colE1 segment of pML21 was digested with restriction endonucleases, electrophoresed on 5% and 10% polyacrylamide gels (0.6 cm $\times$ 12 cm), and the gels were stained with ethidium bromide (0.5 μ g ml⁻¹). DNA bands were visualised in long-wave ultraviolet light and photographed using Kodak No. 23A red filter and Polaroid land pack film type 105. Photographs from 5% gels were shown above and those from 10% gels below. *a-e*, Digests of colE1 DNA; *f* and *g*, digests of the mini-colE1 segment of pML21. Restriction endonucleases used were: (*a*) R·HaeII, (*b*) R·HaeIII, (*c*) R·HaeII + R·HaeIII, (*d*) R·HaeII + R·EcoRI, (*e*) R·HaeIII + R·EcoRI, (*f*) R·HaeII, and (*g*) R·HaeIII. Capitals on the left side of *a*, *d* and *f* are HaeII fragments and those on *b*, *c*, *e* and *g* are HaeIII fragments, except "D" on *c*, that is the HaeII fragment. Roman numerals on the right side show the colE1 DNA fragments created by cleavages with two different enzymes. "X", "Y" and "Z" on *f* and *g* were the fragments found only in the digests of the mini-colE1 segment of pML21. Preparation of colE1 DNA, isolation of R·HaeII, R·HaeIII and R·EcoRI, digestion of DNA and method of polyacrylamide gel electrophoresis have been described⁹⁻¹². The mini-colE1 segment of pML21 was isolated from the EcoRI digest of pML21 DNA by Agarose gel electrophoresis¹².

duced by the cleavages were abbreviated as those by Smith and Nathans⁸. The size of each fragment was expressed as a percentage of the length of the colE1 DNA molecule.

ColE1 and pML21 DNAs were isolated respectively from *E. coli* P678-54 (colE1) and *E. coli* W3350thy-(pML21), as described earlier^{7,9}. R·HaeII and R·HaeIII were prepared from *H. aegyptius* by the method of Roberts *et al.* and R·EcoRI from *E. coli* RY13 by the method of Greene *et al.*¹². Restriction fragments were resolved by electrophoresis on either 5% or 10% polyacrylamide gel¹⁰ or on 1.2% Agarose gel¹², and visualised by staining with ethidium bromide. For isolation of fragments from gels, band regions visualised by a brief staining were homogenised in a few ml of 10 mM Tris-HCl (pH 7.6) and incubated for 15 h at 37 °C. The extract was concentrated and purified by passing through a Sephadex G-100 column (0.4 cm $\times$ 15 cm). The size of HaeII and HaeIII fragments was estimated from the distribution of ³²P in each fragment generated from uniformly labelled ³²P-DNA. The size of other fragments was estimated from the mobility relative to the HaeII and HaeIII fragments.

When colE1 DNA was subjected to complete digestion with R·HaeII, six specific fragments (HaeII-A to -F) were visualised after polyacrylamide gel electrophoresis and subsequent staining of the gels with ethidium bromide (Fig. 1*a*). On the basis of the relative yield of each fragment from uniformly labelled ³²P-DNA, their molecular weights were estimated to be 36.9%, 27.5%, 18.0%, 11.0%, 5.4% and 1.2%, respectively. The order of these fragments was determined from analysis of partial digests followed by redigestion of individual intermediate fragments. A representative electropherogram of partial digests

is presented in Fig. 2, in which several discrete fragments can be seen in addition to the limit digestion products. The approximate size of each band is given by the side of the gel column. DNA banding between HaeII-A and -D was completely digested with R·HaeII and the products were analysed. As a result, it was found that 6 out of 10 bands tested were intermediates of digestion (i1 to i6) and the others were HaeII fragments A, B, C and D. Each intermediate gave limit products as follows: i1 $\rightarrow$ B D, i2 $\rightarrow$ C D F, i3 $\rightarrow$ C E F, i4 $\rightarrow$ C E, i5 $\rightarrow$ C F, i6 $\rightarrow$ D F. By arranging these limit products in overlapping positions, the physical order of the HaeII fragments was deduced to be A E C F D B, with A and B being contiguous in the circular molecule. This order was further confirmed by analysis of the EcoRI site and the constituent HaeII fragments of pML21, as described below.

As one specific break is valuable as a reference point in the circular molecule, the location of the EcoRI cleavage site in the HaeII fragments was determined by comparison between the HaeII digests of the circular colE1 DNA molecule and the EcoRI-treated linear colE1 DNA molecule (EcoRI-A). The only difference in the two digests was the absence of HaeII-A among the EcoRI·HaeII products, and the appearance of two new fragments: one between HaeII-B and -C and the other between HaeII-C and -D (I and II in Fig. 1*d*). The EcoRI site was therefore within HaeII-A. This was confirmed by direct cleavage of HaeII-A with R·EcoRI (data not shown), the two products I and II being identified as EcoRI-A·HaeII-A1 and EcoRI-A·HaeII-A2, respectively. Their sizes were estimated to be 21.4% and 16.4%, respectively. Which end of HaeII-A is closer to the EcoRI site was determined with pML21 as follows.

Table 1 Digestion of purified *Hae* fragments with the second *Hae* enzyme

Original fragment	Products of the second digestion
<i>Hae</i> II-A	<i>HH</i> -II, <i>HH</i> -IV [‡] , <i>Hae</i> III-B, <i>Hae</i> III-F
<i>Hae</i> II-B	<i>HH</i> -I, <i>HH</i> -V [‡] , <i>Hae</i> III-C, <i>Hae</i> III-M*
<i>Hae</i> II-C	<i>HH</i> -III, <i>HH</i> -VII, <i>Hae</i> III-G, <i>Hae</i> III-I, <i>Hae</i> III-K, <i>Hae</i> III-L*
<i>Hae</i> II-E	<i>HH</i> -VI, <i>HH</i> -VIII, <i>Hae</i> III-N
<i>Hae</i> II-F	<i>HH</i> -IX [§] , <i>HH</i> -X [§]
<i>Hae</i> III-A	<i>HH</i> -I, <i>HH</i> -II
<i>Hae</i> III-D [†]	<i>HH</i> -V [‡] , <i>HH</i> -X, <i>Hae</i> II-D
<i>Hae</i> III-E	<i>HH</i> -III, <i>HH</i> -VIII
<i>Hae</i> III-H [†]	<i>HH</i> -IV [‡] , <i>HH</i> -VI
<i>Hae</i> III-J	<i>HH</i> -VII, <i>HH</i> -IX

Each *Hae*II and *Hae*III fragment shown in the first column was purified by polyacrylamide gel electrophoresis and completely digested with the second *Hae* enzyme. Limit products determined by gel electrophoresis are shown in the second column. In parallel experiments, *Hae*II-D, *Hae*III-B, *Hae*III-C, *Hae*III-F, *Hae*III-G and *Hae*III-I were confirmed to carry no *Hae* cleavage site inside.

*The sizes of *Hae*III-L and *Hae*III-M were about equal, one derived from *Hae*II-C was named *Hae*III-L.

[†]Digestion of *Hae*III-D and *Hae*III-H with R-*Hae*II was done in the presence of *Hae*III-C and *Hae*III-G, respectively.

[‡]*HH*-IV and *HH*-V were indistinguishable by gel electrophoresis; one derived from *Hae*II-A was named *HH*-IV. This *HH* fragment should be identical to the one produced from *Hae*III-H, because analysis of overlapping positions at the other end indicates that *Hae*II-A partly overlaps with *Hae*III-H but not with *Hae*III-D. Similarly, the other overlapping fragment produced from *Hae*II-B and *Hae*III-D was named *HH*-V.

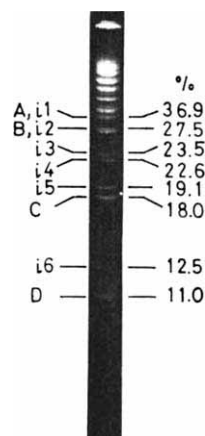
[§]Although not directly demonstrated, *HH*-IX and *HH*-X are evidently derived from *Hae*II-F because the total length of these two fragments is comparable to that of *Hae*II-F and there is no other candidate yielding these fragments.

As previously described⁷⁻¹³, this plasmid replicates as the *colE1* replicon, and yields two fragments when digested with R-*Eco*RI: the longer one contains kanamycin resistance marker but no *colE1* DNA, and the shorter one, defined as the mini-*colE1* segment, continuously carries 49% of the *colE1* genome and a small amount of non-*colE1* DNA sequence (about 5% length of *colE1* DNA). One end of the latter fragment is precisely homologous to one end of the linear *colE1* DNA generated by the cleavage with R-*Eco*RI. Therefore, pML21 should carry one of the two *Eco*RI-A-*Hae*II-A fragments of *colE1*. When the mini-*colE1* segment of pML21 was digested with R-*Hae*II, *Eco*RI-A-*Hae*II-A2 in addition to *Hae*II-C, -E, -F and a new fragment "X" were produced, but not *Eco*RI-A-*Hae*II-A1, *Hae*II-B and -D (compare Fig. 1d and f). The "X" fragment was interpreted to carry a part of *Hae*II-D and the non-*colE1* DNA in the mini-*colE1* segment of pML21 from its electrophoretic

mobility. It was thus concluded that *Eco*RI-A-*Hae*II-A2 was next to *Hae*II-E, confirming that the fragment linked to *Hae*II-E was *Hae*II-A but never *Hae*II-B.

Since use of different restriction endonuclease for fragment analyses at overlapping positions strengthens fidelity of the physical map, digestion of *colE1* DNA with another enzyme R-*Hae*III was performed. R-*Hae*III cleaved *colE1* DNA into 14 pieces (*Hae*III-A to -N in Fig. 1b), in which C and D, G and H, and L and M were respectively inseparable in the conditions used. The molecular weights for *Hae*III fragments (A to N) estimated as in the case of *Hae*II fragments were 17.3%, 16.6%, 14.3%, 14.3%, 6.9%, 6.7%, 6.5%, 6.5%, 4.0%, 2.7%, 1.2%, 1.1%, 1.1%, and 0.8%, respectively. Combined digestion of *colE1* DNA with R-*Hae*II and R-*Hae*III gave 20 pieces as expected from additivity of the number of cleavage sites (Fig. 1c). From comparison between the gel electropherograms of the digests of *colE1* DNA with both R-*Hae*II and R-*Hae*III and with either one, it is evident that 10 out of 20 fragments correspond to *Hae*II-D, *Hae*III-B, -C (or -D), -F, -G (or -H), -I, -K, -L, -M, and -N. This was confirmed by direct digestion of each fragment with the second *Hae* enzyme (see below). As the sizes of *Hae*III-C and -D, and *Hae*III-G and -H were about equal, the fragments carrying no *Hae*II cleavage site were named *Hae*III-C and -G, respectively. It was thus concluded that *Hae*II sites were present in *Hae*III-A, -D, -E, -H and -J, and that all *Hae*II fragments except *Hae*II-D contained *Hae*III substrate sites. The remaining 10 fragments, *Hae*II-*Hae*III-I to -X (designated as *HH*-I to -X), were produced by cleavages with *Hae*II at one end and with *Hae*III at the other end. Their sizes estimated from their electrophoretic mobilities were 9.1%, 8.6%, 5.3%, 3.3%, 3.3%, 2.9%, 1.7%, 1.6%, 0.9% and 0.4%, respectively. To analyse which *Hae*III fragments were contained in a particular *Hae*II fragment, and vice versa, and from which *Hae*II and *Hae*III fragments a particular *HH* fragment was derived, each *Hae*II and *Hae*III fragment except small fragments (that is *Hae*II-F and *Hae*III-K to -N) were purified and digested with the second enzyme, and the fragments produced were determined. The results are summarised in Table 1, from which we can deduce origins of all *HH* fragments and, therefore, relate the two sets of fragments as follows:

Fig. 2 A gel electrophoretic pattern of partial digests of *colE1* DNA with R-*Hae*II. *ColE1* DNA was partially digested with R-*Hae*II by reducing the amount of enzyme, and subjected to electrophoresis on a 5% polyacrylamide gel. Since no partial digestion product with electrophoretic mobility faster than that of *Hae*II-D was found in preliminary experiments, electrophoresis was done at about twice the current of that in Fig. 1 for obtaining good separation. Capitals indicate the band positions of *Hae*II fragments, and i1 to i6 are intermediate fragments of digestion. The approximate size of each band is given on the right-hand side. Since *Hae*II-A and i1, and *Hae*II-B and i2 were respectively too close to isolate, redigestion was performed on the mixture.



<i>Hae</i> II	A	B	D	F	C	E	A
<i>HH</i>	II	I	V	X IX VII	III VII	VI IV	
<i>Hae</i> III	A	(C, H)	D	J	(G, I, K, L)	E	(B, F)

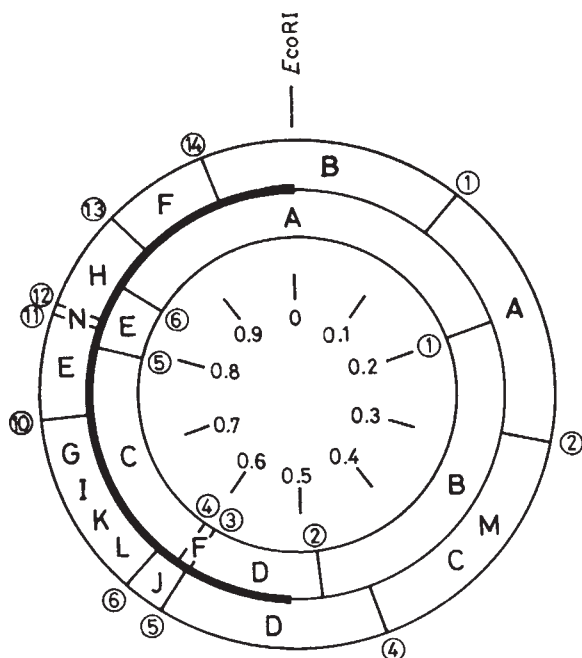


Fig. 3 Physical map of the colE1 genome constructed by the cleavages with R·EcoRI, R·HaeII and R·HaeIII. The inner and outer rings show HaeII and HaeIII fragments, respectively. The EcoRI cleavage site was designated the zero point, and each cleavage site was numbered in the clockwise direction (circled numbers). The map distance from the zero point is shown as the fraction of the length of colE1 DNA in the same direction. Heavy line indicates the region carried by pML21.

To determine the positions of HaeIII-B and -F in HaeII-A, the location of the EcoRI site in HaeIII fragments and the constituent HaeIII fragments of pML21 were analysed. The electropherogram of combined digests of colE1 DNA with R·EcoRI and R·HaeIII indicates that HaeIII-B was split with R·EcoRI into two fragments, EcoRI-A·HaeIII-B1 (11.4%) and EcoRI-A·HaeIII-B2 (5.9%) (I and II in Fig. 1e). Digestion of the mini-colE1 segment of pML21 with R·HaeIII yielded EcoRI-A·HaeIII-B2, HaeIII-E, -F, -G, -H, -I, -J, -K, -L, -N and two new fragments "Y" and "Z" (Fig. 1g). Thus it was concluded that EcoRI-A·HaeIII-B2 and HaeIII-F were within EcoRI-A·HaeII-A2. The appearance of the two new fragments indicates that non-colE1 DNA in the mini-colE1 segment of pML21 contains one HaeIII site.

We can usefully summarise all the foregoing data in a single cleavage map of the colE1 genome, incorporating the various cleavage sites and molecular weight estimates of the fragments (Fig. 3). The EcoRI site was designated the zero point and measurement of the map distance as the fraction of the length of colE1 DNA was made in the direction A B D F C E of HaeII fragments from this point. According to this expression, pML21 carries the region 0.51 to 1.0. Earlier work indicates that replication initiates at 18% distant from the EcoRI site and proceeds towards the distal EcoRI site⁴⁻⁶. The replication origin of colE1 is present in pML21 (ref. 7). Therefore, the origin and direction of DNA replication can be deduced to be 0.82 and anticlockwise on the map in Fig. 3.

We thank Dr K. Sugimoto for helpful discussion and a critical reading of the manuscript.

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Corrigendum

In the article "Prehistoric exploration at Hadar, Ethiopia" by G. Corvinus (*Nature*, **261**, 571; 1976) no acknowledgement was given to the institution which funded the work. The author wishes to thank the Centre National de Recherche Scientifique for a grant in 1974 and for a fellowship in 1975.

Errata

In the article "Reactions involving singlet oxygen anion" by W. H. Koppenol (*Nature*, **262**, 420; 1976) for 'effects', page 420, paragraph 1, line 6, read 'involvement'. Also, page 421, line 29, insert 'quantitatively' between 'reacts' and 'with'.

In the article "New cranium of *Homo erectus* from Lake Ndutu, Tanzania" by R. J. Clarke (*Nature*, **262**, 485; 1976) the following corrections should be made. In the first paragraph on page 485 humanoid should read hominid. The semicolon in the third line of the second paragraph on page 486 should be replaced by a comma and the last word in the same line should be 'lacking'. The last sentence in the same paragraph should read 'In addition, there are the isolated right tympanic plate and both isolated petrous temporals'. There is no scale on Fig. 1: the distance from a to O is 12 cm.

In the article "RNA polymerase specificity and the control of growth" by A. Travers (*Nature*, **263**, 641; 1976) the following corrections should be made on page 643: Paragraph 3, the sentence beginning in line 16 should read '... addition of T7 DNA results in a reduction of T2 RNA synthesis and a stimulation ...'; and the sentence beginning in line 28 should read '... the relative competition of T2 RNA and rRNA synthesis is virtually independent of the nature of the competing template'; and in paragraph 6, the second sentence should read 'In two instances where stable RNA synthesis is shut off *in vivo*, ppGpp accumulation and T4 infection ...'.

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matters arising

Asymmetric degradation of DL-leucine with longitudinally polarised electrons

BONNER *et al.* reported that when DL-leucine was irradiated with natural, antiparallel, polarised electrons of 120 keV energy, D-leucine was decomposed to a greater extent than L-leucine: with unnatural, parallel, polarised electron irradiation, however, L-leucine was degraded to a greater extent¹.

The mechanism of this phenomenon is not known. The authors were unable to decide whether the asymmetric degradations was caused by the longitudinally polarised electrons themselves, or by their bremsstrahlung. I wish to point out that the degradations measured could not have been caused by the bremsstrahlung of the bombarding electrons.

The fact that $\Delta E = 29$ eV for three cases is remarkable and shows that the law of exponential decomposition is applicable in the analysis. The value of ΔE coincides with the same values in the case of bremsstrahlung irradiation³. The ΔE value is very reasonable, showing that there is no need to assume any chain-type reaction mechanism.

The values of relative difference in the decomposition rates ($2Z$) show a broader spread. This, and the large value of ΔE in experiment (2), can be understood by taking into account the remarks of Bonner *et al.*, that it was difficult to control many of the experimental parameters of the polarised electron source. Even so, I feel that the value $2Z \approx 10\%$ can be accepted. (The upper limit for the same quantity in the case of DL-alanine was $2Z \approx 0.1\%$ (ref. 4).)

The energy of the incoming electrons

$2Z \approx 10\%$. A mechanism based on the direct interaction of polarised electrons with the asymmetric molecules, which was first outlined in ref. 6 and needs to be worked out in detail, may provide an approach to the understanding of asymmetric degradation.

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Table 1 Analysis of the asymmetric degradation of DL-leucine

Experiment	Irradiation nA h	Sample decomposition	Energy/decomposed molecule (eV)	$\frac{\epsilon_D - \epsilon_L}{\epsilon_0}$	Weighted average
(1)	331	52.9	29.3	0.189 ± 0.039	0.151 ± 0.024
(2)	498	50.9	55	0.129 ± 0.030	
(3)	795	73.9	29	0.085 ± 0.015	0.081 ± 0.012
(4)	860	75.6	29	0.074 ± 0.020	

Analysing the data given in Table 1, of ref. 1, I calculated the average energy spent by the electrons to decompose a single leucine molecule (ΔE) and the relative difference of the decomposition rates for L-, and D-leucine ($2Z = (\epsilon_L - \epsilon_D)/\epsilon_0$). For this purpose the following simple equation was used (which assumes an exponential decomposition of the molecules under constant irradiation²)

$$\frac{N_L - N_D}{N_L + N_D} = PZ \ln \frac{1}{1 - D} \quad (1)$$

where the left side is the relative difference in the number of remaining L and D molecules, that is, the enantiomeric selectivity ($\%L - \%D$) defined in ref. 1; P is the polarisation of the incoming electron beam (percentage excess in Table 1 of ref. 1); and D is the sample decomposition (percentage divided by 100). The thickness of the irradiated DL-leucine samples (~ 20 mg cm⁻²) was nearly equal to the range of the bombarding electrons.

The data are presented in Table 1.

is lost in the sample by ionisation and radiation; the latter is the bremsstrahlung.

The fraction of the incident energy which is converted into bremsstrahlung in a thick target is approximately⁵

$$I/E = 7ZE \times 10^{-4} \quad (2)$$

where Z is the atomic number of the bombarded material (~ 7) and E is the energy of electrons in MeV.

Even if the whole energy of the bremsstrahlung had been absorbed in the sample, less than 0.1% of the degraded molecules would have been decomposed by bremsstrahlung. If the low degree of polarisation of the low energy bremsstrahlung ($P \approx 10^{-3}$) and the very low upper limit for Z ($< 3 \times 10^{-3}$ according to the measurements in ref. 2) were also taken into account, the upper limit of the enantiomeric selectivity would be $\sim 10^{-6}\%$. It is not possible therefore to understand the great difference found by Bonner *et al.* if the asymmetric degradation is caused by bremsstrahlung. There is no known mechanism to explain the effect of

BONNER ET AL. REPLY—Keszthelyi calculates that the average energy spent by the electrons to decompose a single leucine molecule is about 29 eV, that less than 0.1% of the degradations were due to bremsstrahlung, and that therefore ionisation must have engendered $> 99.9\%$ of the degradation as well as (in some unspecified manner) the observed asymmetric effect. We wish to comment on Keszthelyi's interpretation¹ of our results.

Using the sample weight, percentage degradation and total coulombs used in our experiments², one readily calculates that more than 3,000 molecules of DL-leucine were decomposed for each impinging primary electron. If each leucine decomposition requires an average 29 eV (ref. 1), each primary 120-keV electron thus loses an average total of about 87 keV in decomposing the leucine substrate. The remaining 32 keV presumably is lost in decomposing the colloidal binder and in the rather inefficient geometry of the leucine target. There are thus some 3,000 primary processes initiated by each incident chiral electron. If primary ionisation is the predominant chiral process, the chiral efficiency of each such event must on average approach 5% to explain the asymmetric effect observed. If, as is possible, the incident electron loses its chirality after fewer collisions, the chiral efficiency must be still greater. What mechanisms might permit this?

Keszthelyi is correct that $< 0.1\%$ of the chiral primary electron energy is available as bremsstrahlung, which would

be fully circularly polarised only at the low intensity³, high energy end of its spectrum⁴. Thus it would seem unlikely that circularly polarised bremsstrahlung alone could explain our asymmetric effect² (ignoring such unknown amplification mechanisms as might involve the crystal lattice). There is, however, an additional source of chiral electromagnetic radiation potentially available for asymmetric photolytic decomposition—circularly polarised phosphorescence or fluorescence radiation emitted by the chiral substrate after excitation by the chiral primary electrons⁵. Another alternative amplification mechanism might involve chiral secondary electrons of about 29 eV, liberated through ionisation of the chiral substrate molecules by chiral primary electrons. Such chiral secondary electrons should be quite abundant, that is, about 3,000 per initial chiral primary electron. Experiments which we hope may shed light on these questions are in progress.

Our work was supported in part by the Office of Naval Research, NASA and ERDA. We are grateful to Professor H. P. Noyes (Stanford Linear Accelerator Centre) for his contributions to our reply.

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Fractures and movement in the Adelaide rift zone

FOLD patterns in the Adelaide geosyncline have been interpreted by Coward¹ as evidence of left-lateral shear between two Australian subplates, although the crustal shears have not been defined in detail. It has been implied further that left-lateral movement still exists in this region^{1,2}. Apparently, studies of earthquakes^{3,4} have not been taken into account in the analysis of the tectonics of South Australia, since these indicate that right-lateral shear occurs at present, and may have done so in the past.

The seismicity data quoted¹ contain few events³, and only show that seismic activity exists in the region of the rift zone. More recent data suggest that well-defined trends in activity occur³, which may outline the main fracture zones in the crust (Fig. 1). From focal mechanisms⁴ and trends in energy release⁶, we propose the tectonic model shown in Fig. 2. This is modified from Stewart and Mount⁴, and may be further altered for the eastern and southern extensions of the rift zone. Present faulting varies from strike slip near Adelaide to dip slip in the area of the Flinders Ranges (Fig.

2), and is more complex than the simple shear model used to interpret folding¹. The lateral extent of the seismicity trends (Fig. 1) and focal depths³ suggest that the shear zones in Fig. 2 represent fractures extending to the lower crust, and possibly into the upper mantle. The proposed shear zone at the northern end of the rift zone coincides with a region of high conductivity in the deep crust and upper mantle⁷ (Fig. 2). Since such shear zones may be active over extended periods of geological time⁸, the boundaries defined in Fig. 2 may represent the major lines of weakness in the crust throughout the life of the geosyncline (that is, since the late Proterozoic). The shear zones in Fig. 2 are approximately parallel to the sinistral shear couple proposed by Coward¹.

of the Adelaide rift zone (Fig. 2) may be compatible with the north-south compressive stress observed in central Australia⁹.

Widespread diapirism in the northern section of the rift zone (Fig. 2) seems to have occurred in conditions of regional tension (T. J. Mount, personal communication). On the model of Fig. 2, this implies that right-lateral movement occurred between the subplates at the time of diapirism. The intrusion of Proterozoic carbonate material into the overlying cover rocks may have continued from the Upper Proterozoic at least into the Cambrian¹⁰. Thus although left-lateral movement may have occurred at various times in the past, this may not always have been the case, and is probably not so at present. Rather, the movement may be

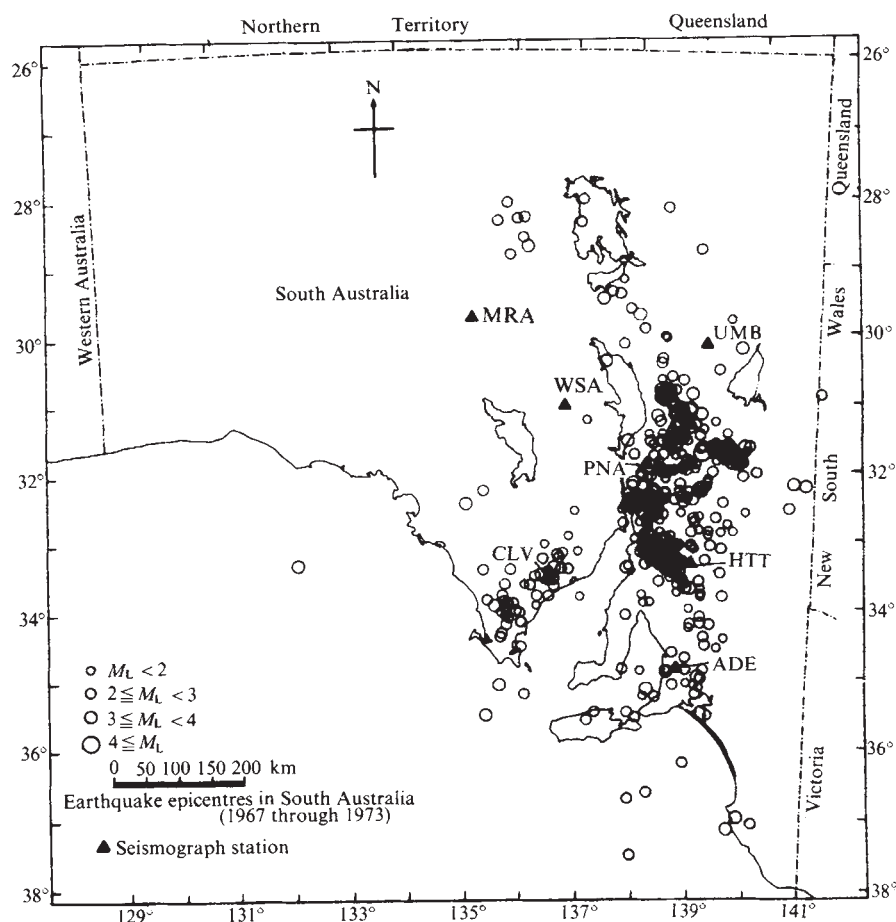


Fig. 1 South Australian seismicity, 1967–73.

Mechanism studies⁴ strongly suggest that right-lateral movement occurs between the eastern and western sections of the system, unlike the left-lateral shear originally proposed by Cleary and Simpson² and quoted by Coward¹. The right-lateral movement may continue southward along the transform fault to the mid-ocean ridge. This sense of motion on the north-western extension

oscillatory⁴, perhaps because of variations in plate velocity across Australia. The postulated left-lateral movement of 150 km (ref. 1) may be large in view of the remarkable uniformity of crustal thickness over much of the Adelaide Rift Zone¹¹. Although this may have been possible if faulting was almost pure strike-slip (as on the northwestern extension of the rift), smaller relative

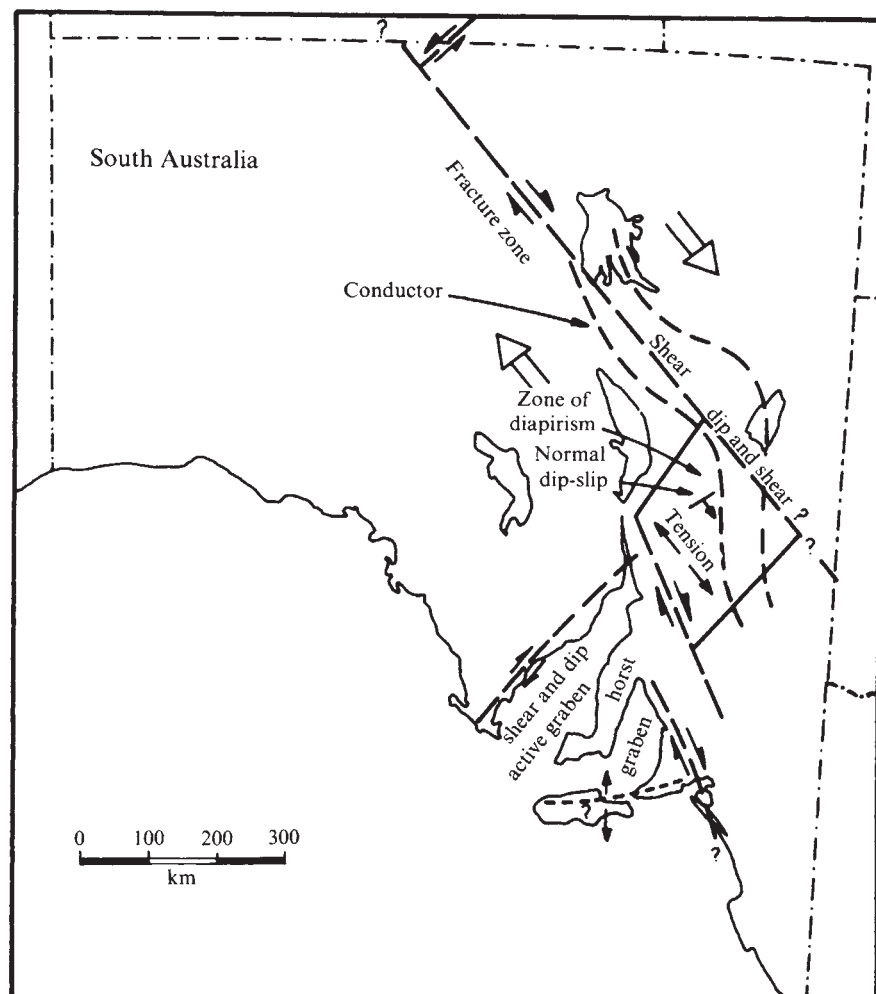


Fig. 2 Tectonic model for South Australia, showing the present sense of movement.

movements of only a few tens of kilometres may have been sufficient to produce vertical movements of several kilometres and much of the known geology of the region¹.

Right-lateral shear may have been just as important as left-lateral movement in the Adelaide Rift Zone, while dip-slip faulting may have been prominent in parts of the geosyncline. An interpretation of fold patterns should therefore allow for possible variations in the orientation, style and sense of faulting. The extent of shear movement across the whole system may also need revision.

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Large scale Palaeozoic shear zone in Australia

HYPOTHESES invoking large scale horizontal displacements¹, fracture zones and the movement of plates², then ideas rejecting 'megashield'³, and now a revival of the large scale Palaeozoic shear zone concept⁴ have been generated to explain the pattern of geological structures observed in the Adelaide geosyncline of South Australia. The latest attempt⁴ has curiously ignored reference to earlier discussions¹⁻³ which I believe are strongly relevant to the basic argument of whether a Palaeozoic shear zone is a tenable hypothesis. It would seem that the whole spectrum of concepts has now been covered; sinistral movements⁴, dextral movements¹ and no relative movement³. Although Coward's case⁴ is rather difficult to assess, the idea seems to have appealed to Sutton⁵ who cited Coward's model as a possible example of deformation within continental plates.

Crawford and Campbell's model¹ supposed that, "The Adelaide geosyncline originally extended meridionally along the line of the present Flinders Ranges . . .", whereas on Coward's model⁴, "a regional pattern of folds on north-east-south-west axial planes was modified by a sinistral shear couple . . .".

Palaeomagnetic evidence from the Precambrian of South Australia was cited⁶ to disprove the extension of the Crawford-Campbell model to involve regions of the Gawler Block; this has since been acknowledged⁷. We made it quite clear, however, that the data obtained earlier from the southern part of the Adelaide geosyncline⁸ were inadequate (because of metamorphic effects), to permit comment on whether that region had suffered 'rotation' from shearing. The only region of the Adelaide geosyncline to yield acceptable palaeomagnetic data was the central region of the Flinders Ranges⁹. Those data, by comparison with late Precambrian-Ordovician data from Western Australia, central and northern Australia, are consistent with the principal axis of the geosyncline having maintained a meridional trend. The data are not consistent with originally north-east-south-west trending axial planes.

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Matters arising

Matters Arising is meant as a vehicle for comment and discussion about papers that appear in *Nature*. The originator of a Matters Arising contribution should initially send his manuscript to the author of the original paper and both parties should, wherever possible, agree on what is to be submitted. Neither contribution nor reply (if one is necessary) should be longer than 300 words and the briefest of replies, to the effect that a point is taken, should be considered.

obituary

Keith Bullen came to Cambridge from New Zealand, and entered St. John's College in 1931. It was his great good fortune there to meet Harold Jeffreys, who was just embarking on a task which was to be momentous for the development of seismology. This was the construction of new travel-time tables for earthquake pulses, using sophisticated statistical techniques and taking full advantage of the data collected over several years in the International Seismological Summary. Jeffreys asked Bullen to assist him. In spite of the very irregular distribution of seismological stations over the Earth's surface and the inaccuracies of instruments at that time, the Jeffreys-Bullen Tables (1935 and 1940) have endured astonishingly, and are still in common use.

In 1936, using the distributions of seismic velocities with depth which were derived by inverting the travel times, Bullen obtained his first model of the distribution of density throughout the Earth—and for the rest of his life he was deeply involved in successive efforts to improve density models. He told the story of his work with skill and enthusiasm in his book *The Earth's Density*, which was published in 1975.

Bullen explored the density distribution in two ways. The seismic velocities alone are not sufficient to determine the two elastic constants and the density. One further physical assumption is

needed. In the series of models called *Bullen A* he used a relationship between pressure, density and compressibility following the method of Adams and Williamson, whereas in the series *Bullen B* he assumed that compressibility and the rate of change of compressibility with regard to pressure varied smoothly throughout most of the Earth's interior. The results of the two different methods were largely reconciled when satellite observations produced a significant correction in the accepted value of the Earth's moment of inertia.

After 1960, when observations of the periods of the free oscillations of the Earth became available, Earth models could be revised so as to fit these also. Bullen and Hadden produced a new series of models designed to do that, but Bullen noted that they had not been able to allow for the effect of anelasticity, which, as Jeffreys had pointed out, tended to lengthen the periods of free oscillations.

His experiences in trying to compare Earth models put forward by different workers convinced Bullen of the need for a single reference model with which all variants could be compared. He became Chairman of the Committee for a Standard Earth Model, sponsored by the I.U.G.G., and worked hard for it until ill health intervened.

Bullen wrote an *Introduction to Seismology* which is uneven in its demands on the reader, but has run

into three editions and is still found very valuable. He also wrote an excellent *Introduction to Dynamics*, now in its eighth edition. A great asset of the book is Bullen's effort to give the reader insight into the reasons which determine the next step to be taken, and to explain why certain methods succeed and others fail. Throughout the book one finds, under the heading S.P.I.C.E. (special point(s) in choosing examples) the type of helpful suggestions that a good tutor would make to a pupil.

Bullen was a brilliantly clear expositor, whether writing or lecturing. He was also a very effective and strong (sometimes obstinate) committee man, so that he was called to many positions of responsibility, especially on Australian scientific bodies and in the I.U.G.G. and the International Association of Seismology and Physics of the Earth's Interior. His outstanding contributions to seismology were widely appreciated, and many honours fell to him, including Fellowship of the Royal Society in 1949. He was awarded the Gold Medal of the Royal Astronomical Society, and medals of several Australian and American scientific societies.

Bullen was a man of great energy and resource, in spite of partial deafness throughout much of his working life he travelled very widely. He died on September 23, 1976, aged 70, leaving colleagues and friends in every continent.

E. R. Lapwood

announcements

Meetings

November 19, **Magnetospheric Dynamics**, London (Royal Astronomical Society, Scientific Societies Lecture Theatre, 23 Savile Row, London W1, UK).

November 23–26, **Future of Aircraft All-Weather Forecasting**, London (The Manager, Conference Department, Institution of Electrical Engineers, Savoy Place, London WC2R 0BL, UK).

December 6–10, **Fall Meeting of the American Geophysical Union**, San Francisco (Meetings Registration, American Geophysical Union, 1909 K Street, N.W., Washington, D.C. 20006).

December 9–10, **Technologies for Rural Health**, London (The Executive Secretary, The Royal Society, 6 Carlton House Terrace, London SW1Y 5AG, UK).

March 30–April 1, **Mixing**, Cambridge (The Organising Secretary, 2nd Mixing Conference, BHRA Fluid Engineering, Cranfield, Bedford MK43 0AJ, UK).

September 13–15, **Particle Size Analysis**, Salford (Dr D. Dollimore, Department of Physical Chemistry, University of Salford, Salford M5 4WT, UK).

October 25–28, **Radar**, London (Deadline for abstracts: January 10) (IEE Conference Department, Savoy Place, London WC2R 0BL, UK).

November 22–24, 1977, **Design and Applications of EHV Substations**, London (Deadline for abstracts: November 29) (IEE Conference Department, Savoy Place, London WC2R 0BL, UK).

April 16–20, 1978, **Luminescence**, Salford (D. Irish, Pye Unicam Ltd., York Street, Cambridge CB1 2PX, UK).

September 4–7, 1978, **Medicinal Chemistry**, Brighton, Sussex (Symposium Secretariat, VIth International Symposium on Medicinal Chemistry, 31 Plane Tree Way, Woodstock, Oxford OX7 1PE, UK).

September 18–22, 1978, **Fat Research**, Brighton, Sussex (The Assistant Secretary, S.C.I., 14 Belgrave Square, London SW1X 8PS, UK).